THE UNITED STAMENS OF RUBIACEAE. MORPHOLOGY, ANATOMY; THEIR ROLE IN POLLINATION ECOLOGY[1]

Christian Puff, Anton Igersheim, Ralf Buchner, and Ulrike Rohrhofer[2]

ABSTRACT

Based on morphological and anatomical investigations of selected species of *Argostemma*, *Steenisia*, *Strumpfia*, *Acranthera*, *Antherostele*, and *Posoqueria*, and reports in the literature about several other taxa, the paper surveys how unions in the androecium of Rubiaceae can come about. In *Argostemma*, the formation of anther tubes is either by union of adjacent apical connective appendages and agglutination of neighboring pollen sac walls, or only by union of connective appendages (with or without sterile remnants of pollen sacs); the anthers dehisce longitudinally and the subepidermal cell layer (endothecium) has thickenings. *Argostemma*, moreover, has species without proper anther tubes in which the anthers (with apical pores and a pollen sac wall without thickenings in the subepidermal cell layer) are, nevertheless, variously grouped together. The anther tube of *Steenisia* comes about by the interlocking of epidermal layers of neighboring apical connective appendages and agglutinated cuticles. *Strumpfia* is unique in having a special cell layer uniting all anthers. The strictly cylindrical anther tube of *Antherostele* is, like in the Asteraceae, held together by agglutinated cuticles of neighboring pollen sacs; apical connective appendages are not involved in the tube formation. *Acranthera* has an "anther-style and stigma complex" (stamens, united by apical connective appendages, are firmly connected with the stigma; descriptions and comments about the highly unusual stigmatic areas in this genus are also included). *Posoqueria* has an anther head, which is held together only by agglutinated, sterile appendages or extensions of the thecae that are produced both basally and apically. The unstable nature of the anther complex is a prerequisite for proper function of *Posoqueria*'s unique "catapult mechanism" of the stamens, by which pollen is hurled at the pollinator. *Argostemma*, *Steenisia*, *Neurocalyx*, and *Strumpfia* are believed to be buzz pollinated. In taxa with longitudinal anther dehiscence (*Argostemma* in part, *Steenisia*, *Neurocalyx*), the apically constricted anther tubes with porelike openings are thought to represent structures analogous to the poricidally dehiscent anthers of buzz-pollination systems. It is speculated that at least some *Acranthera* species may also be buzz pollinated; the windows formed by the elongated connective appendages above the longitudinally dehiscent anthers might take over the function of pores.

Agglutinations or fusion in the androecium, or unions by other means, are not confined to the "Synandrae" (= Asteridae sensu stricto) but also occur in various other families, including a number of Rubiaceae. In one rubiaceous genus, there is even an "anther-style and stigma complex." These phenomena are often imprecisely and vaguely described as "anther tube," "anther cone," "anthers convergent," "anthers connivent," or "anthers connate."

One of the aims of the present paper is to show the range of variation of the "synandry" syndrome in the Rubiaceae, which apparently has evolved independently in various alliances. Detailed morphological and anatomical documentation, based on the investigation of preserved material and supplemented by field observations, will be presented for selected taxa.

The presence of united anthers ("united" is used here in a neutral and wide sense, i.e., presence of agglutinations, proper fusion, or union by other means in the androecium), as well as shape, size, color, presentation, and orientation of the entire androecium, appears to be intimately linked with pollination biological specializations.

MATERIALS AND METHODS

Detailed morphological and anatomical studies were carried out on the following taxa and collections (material preserved in 70% alcohol or FPA; vouchers deposited in WU, unless stated otherwise). Methods used for SEM (scanning electron microscope) investigations and microtome techniques are described in Puff et al. (1993) and Igersheim (1993a). **: carried out on reconsti-

tuted herbarium material (for details on method see Buchner & Puff, 1993).

Acranthera Arn. ex Meisn. (Isertieae?, incertae sedis): *A. atropella* Stapf: Borneo. Sabah: Tawau Hills Park, *Puff & Buchner 920419-1/4* (Fig. 11; 11A from color slide Puff SEA-1887). *A. frutescens* Val.: Borneo. Brunei: Temburong Distr., *Puff & Buchner 920506-1/1* (Fig. 10; 10A from color slide Puff SEA-1643). *A. involucrata* Val.: Borneo. Brunei: Temburong Distr., *Puff & Buchner 920506-1/2* (Figs. 8, 9A–E; 8A from color slide Puff SEA-1649). *Acranthera* sp., aff. *A. variegata* Merr. (subg. *Dichroanthes* Bremek.): Borneo. Brunei: Temburong Distr., *Puff 900818-1/9* (Fig. 9F, G).

Antherostele Bremek. (Urophylleae): *A. banahaensis* (Elmer) Bremek.: Philippines. Luzon: Sorsogon Prov., Mt. Bulusan, *Elmer 16166* (US)**. *A. grandistipula* (Merrill) Bremek.: Philippines. Luzon: Camarines, *Sulit B.Sc. 30316* (A)** (Fig. 12).

Argostemma Wall. (Argostemmateae): *A. borragineum* Bl. ex DC.: Borneo. Sabah: Crocker Range, *Puff 890705-1/3* (Figs. 1, 2). *A. diversifolium* Ridl.: Thailand. Peninsula, Trang Prov., forests behind Kachong Bot. Garden, *Puff 900910-1/6* (Fig. 5); Surat Thani Prov., just outside Khao Sok National Park, *Puff & Sridith 930705-1/1* (Fig. 4C, from color slide Puff TH-1915). *A. moultonii* Ridl.: Borneo. Sabah: Crocker Range, Mt. Alab, *Puff 890705-1/2* (Fig. 4A, from color slide Puff SEA-125). *A. neurocalyx* Miq.: Thailand. Peninsula, Krabi Prov., Khao Phanom Distr., Khao Phanom Bencha National Park, *Puff & Sridith 930731-1/2* (Fig. 6A–C, G, H); Surat Thani Prov., Khao Sok National Park, *Puff & Sridith, 930703-1/7* (Fig. 6D–F, I). *A. pictum* Wall.: Thailand. Peninsula, Surat Thani Prov., just outside Khao Sok National Park, *Puff & Sridith 930705-1/2* (Fig. 4D, from color slide Puff TH-1872). *A. psychotrioides* Ridl.: Borneo. Brunei: Belait Distr., *Puff 900807-1/1* (Fig. 3B), 900810-1/9 (Fig. 3A, C–F). *Argostemma* sp.: Thailand. Peninsula, Phattalung Prov., Ban Phut Distr., Khao Bantad Wildlife Sanctuary, Pri Wan waterfall, *Puff & Sridith 930725-1/1* (Fig. 4B, from color slide Puff TH-2220).

Posoqueria Aubl. (Gardenieae subtribe Gardeniinae): *P. latifolia* (Rudge) R. & S.: Origin unknown, cult. Indonesia, Bot. Garden Bogor, *Puff 920913-1/s.n.* (Fig. 13, from color slides Puff SEA-2214, SEA-2216); cult. National Bot. Garden Belgium sub 64-0515, as *P. panamensis* (Walp. & Duchass.) Walp. (Fig. 14).

Steenisia Bakh. f. (Rondeletieae): *S. pleurocarpa* (Airy Shaw) Bakh. f.: Borneo. Brunei: Temburong Distr., *Puff 900818-1/12* (Fig. 7; 7A from color slide Puff SEA-1276).

Strumpfia Jacq. (incertae sedis): *S. maritima* Jacq.: Cuba. Santiago de Cuba, between Daiquiri and Siboney, *Igersheim 920603-1/1* (Fig. 4E, from color slide Igersheim CU-92/78).

Non-rubiaceous taxa: *Ardisia* sp. (Myrsinaceae): Borneo. Brunei: Temburong Distr., *Puff 920506-1/s.n.* (Fig. 4F, from color slide Puff SEA-1657). *Solanum* sp. (Solanaceae): Colombia. Los Nevados Nat. Park, *Puff 920220-1/s.n.* (Fig. 4G, from color slide Puff CL-201).

RESULTS

ARGOSTEMMA (FIGS. 1–3, 4A–D, 5, 6)

Argostemma is a relatively large genus (ca. 150 spp.) with diverse floral characteristics. Corolla shapes vary from the "common" subrotate corollas to bell-shaped types. The anthers are either dehiscent longitudinally or open by means of apical pores. In the former group, the anthers typically have (long) apical connective appendages, and the anthers form a solid tube. In taxa with poricidal dehiscence the anthers may or may not be connate, and apical connective appendages are absent.

Comprehensive investigations have so far not been available for any *Argostemma* species, and the present investigation must restrict itself to a few selected examples. Thus data here presented may represent only a small, possibly incomplete selection of the diverse situation in the genus.

Species with longitudinal dehiscence of the pollen sacs and anthers united into a tube, as exemplified by A. borragineum *and* A. psychotrioides.

Argostemma borragineum has a straight anther tube, and its general flower shape is similar to that of *A. moultonii* (Fig. 4A). The anther tube is distinctly 5-ridged due to the presence of prominent, rounded connectives, especially from the upper
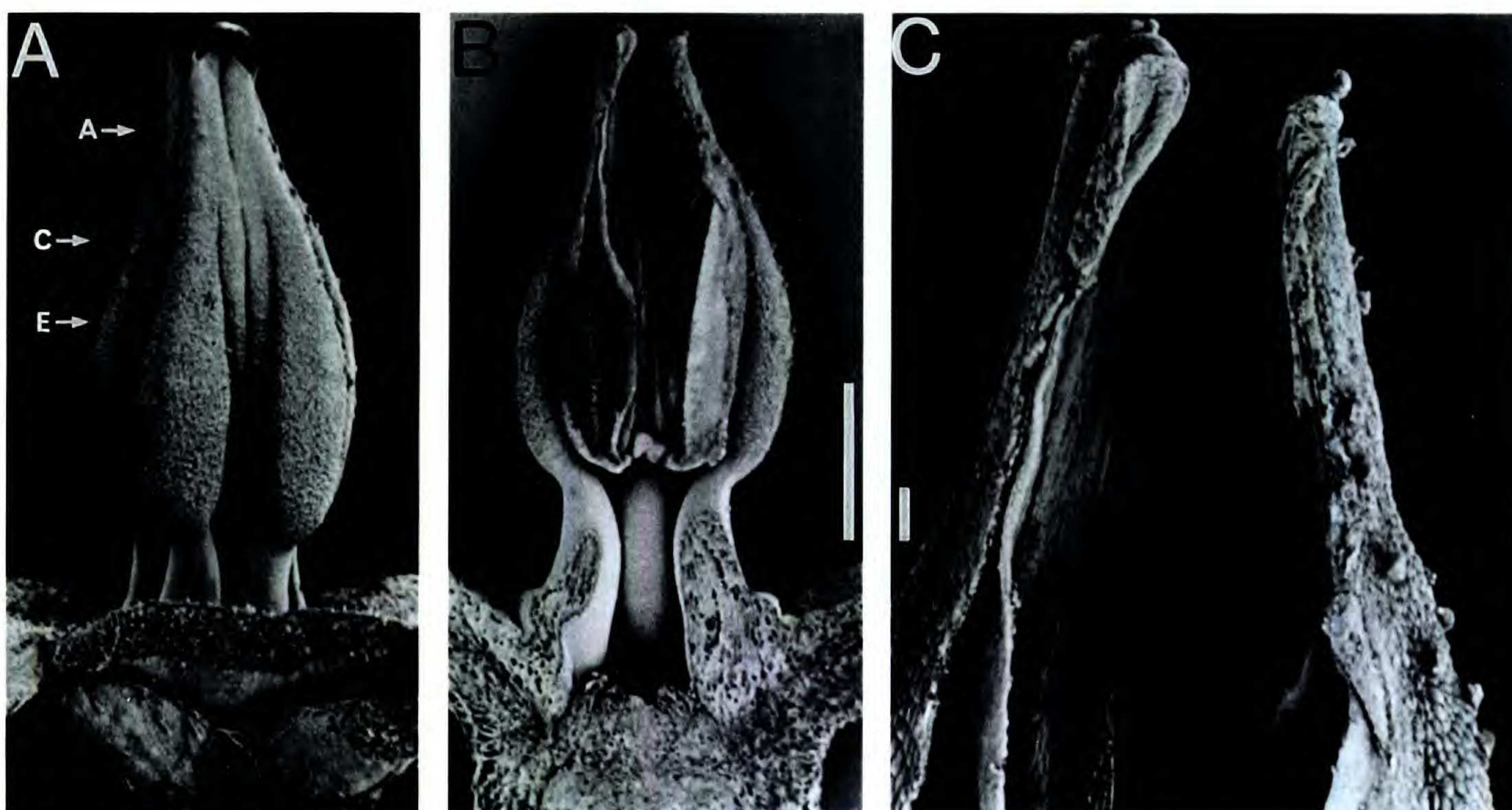
Figure 1. *Argostemma borragineum.* —A. Calyx and corolla removed to show anther tube; the arrows indicate the approximate levels of the sections reproduced in Figure 2.—B. Section of anther tube to show longitudinal dehiscence of pollen sacs and apical appendages.—C. Detail of upper part of anther tube; note free ends of apical connective appendages and upward extensions of pollen sacs. SEM-graphs. Scale bars: 1 mm (A = B); 0.1 mm (C).

third of the anther tube downward (Figs. 1A, 2E, F). The sequence of sections reproduced in Figure 2C–F shows the shape change of the connectives and the pollen sacs. The filaments are very short and somewhat broadened near the base.

The apical part of the anther tube is comprised of sterile anther appendages. The majority of the volume of these apical appendages is made up of prolonged connective tissue, with a distinct "inner" and "outer" epidermis and some layers of parenchymatic tissue between (Fig. 2B). On either side of each angled connective appendage are sterile pollen sac extensions (Fig. 2A, also cf. Fig. 1B, C). These sterile upward extensions of the pollen sacs appear to reach the apex of the anther tube, but they should not be mistaken for dehiscent pores. On the contrary, the pollen sacs open by means of longitudinal slits, and pollen is released into the interior of the anther tube (Figs. 1B, 2D–F).

The entire anther tube forms a solid, united structure. It is always the epidermal layer of pollen sacs from neighboring anthers, which, over a restricted, narrow area, are connected with each other by their fused cuticles (Fig. 2), even in the uppermost, sterile part of the tubes. Detailed study of the pollen sac wall reveals the presence of an endothecium with ringlike thickenings (Fig. 2G).

The fine structure of the anther tube of *Argostemma psychotrioides* differs from that of *A. borragineum* in certain details:

The apical parts of the anther tube are comprised of connective appendages only; there are no sterile extensions of pollen sacs (Fig. 3A). The apical appendages are united in a different way than in *A. borragineum*: The epidermis of the connective appendages develops elongated outgrowths, which are uniseriate but often two- or even three-celled. These outgrowths of neighboring connectives are interlocked, and in between them there is always an intensively stained, mucilaginous substance (Fig. 3B).

The section shown in Figure 3C goes through the uppermost parts of the pollen sacs, which already contain pollen grains. Still, the connection of neighboring anthers by means of interlocked elongated, fingerlike epidermis outgrowths is the same as higher up on the anther tube.

Nearer to the base of the anther tube (Fig. 3D), these specialized cells are no longer present. The epidermal cells of pollen sacs of neighboring anthers are merely in close proximity to each other, but an obvious unification is not detectable (Fig. 3E). As in *A. borragineum*, the endothecial layer shows ringlike thickenings. Tangential sections (Fig. 3F) show that the shapes of the endothecium cells vary from more or less rectangular to fusiform within the same pollen sac wall. It is hardly possible to correlate cell shape and the kind of thickening with the four types of endothecial cells Bremer (1989) distinguished in Bornean *Argostemma* species.
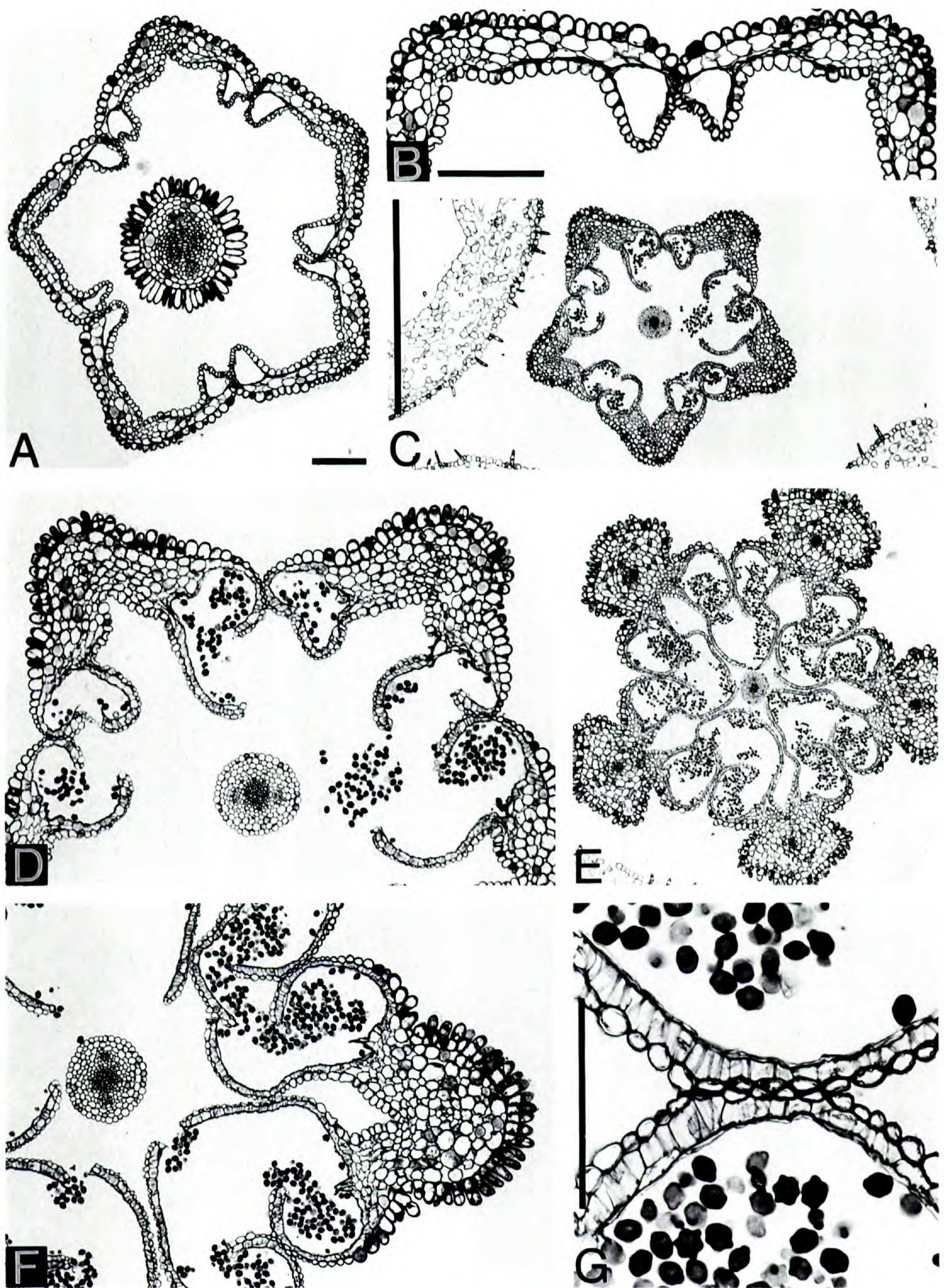
Figure 2. *Argostemma borragineum*, cross sections of the anther tube.—A. Apical connective appendages with a sterile pollen sac remnant on either side, the stigma in the center (elongated peripheral cells: stigmatic papillae).— B. Detail of A, showing agglutination of adjacent connectives and sterile, terminal portion of pollen sacs.—C. Above the middle of the anther tube; as compared to lower down (cf. E), pollen sacs still small in size.—D. Detail of C, pollen sacs of adjacent anthers agglutinated; note shape of connectives.—E. Approximate middle of anther tube; note prominent, dorsally rounded connectives.—F. Similar to E; note dehisced pollen sacs.—G. Agglutinated pollen sacs
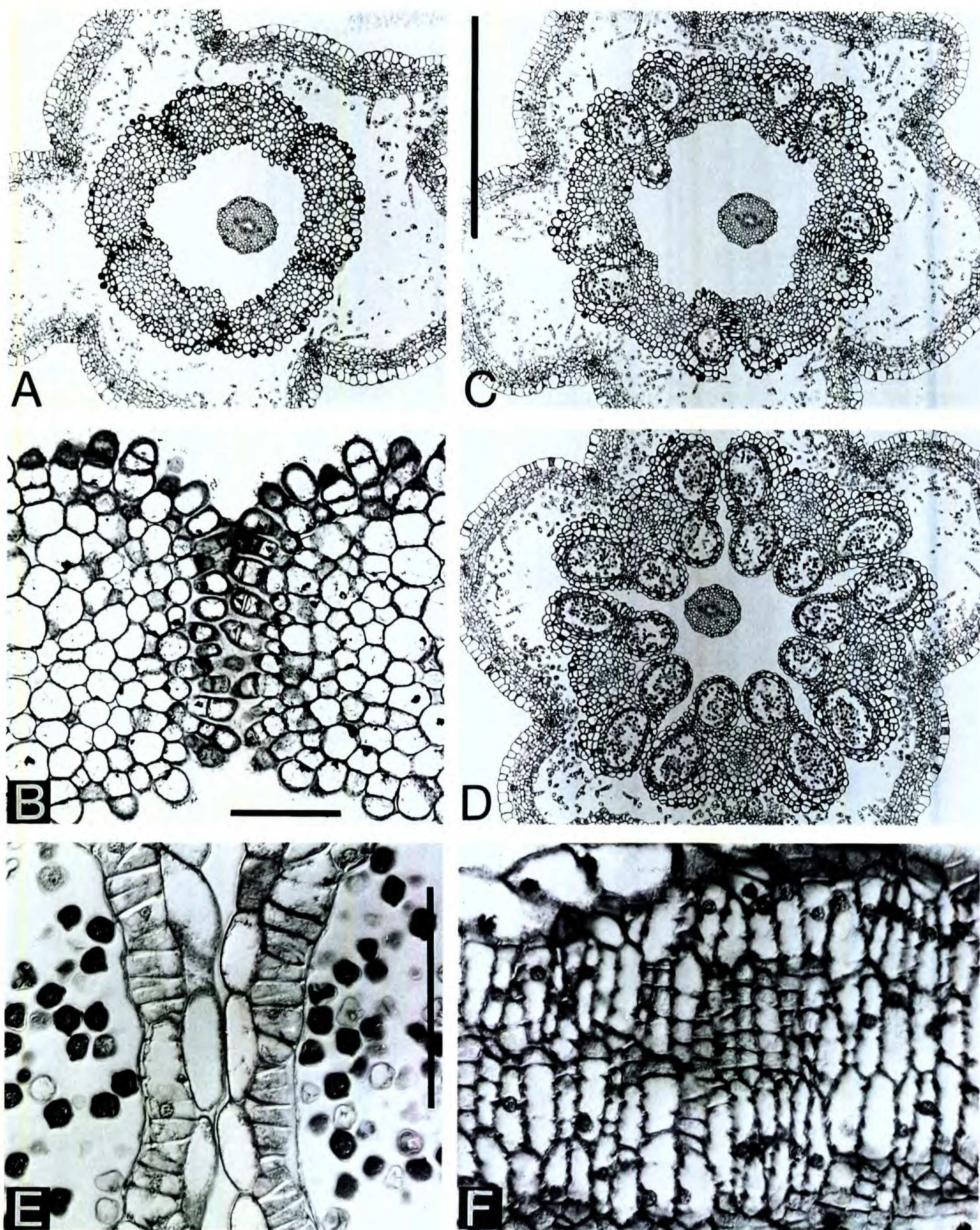
Figure 3. *Argostemma psychotrioides.* A–E, Cross sections of flower buds shortly before anthesis. — A. Uppermost part of anther tube; ring formed by apical connective appendages (corolla outside; style in the center). — B. Interlocked cells of two adjacent connectives. — C. Lower level than A, the uppermost parts of the pollen sacs are visible on either side of the connective. — D. Approximate middle of anther tube; note unequal size of adaxial and abaxial pollen sacs. — E. Detail of pollen sacs of two adjacent anthers, no obvious agglutination is discernible; note thickenings in the endothecium. — F. Tangential section of endothecium. A–F: LM-graphs, E–F: Differential Interference Contrast. Scale bars: 1 mm (A = C = D); 0.1 mm (B; E = F).

←

of adjacent anthers; note thickenings in the endothecium. A–G: LM-graphs (light micrographs). Scale bars: 1 mm (C = E); 0.1 mm (A = D = F; B: G).
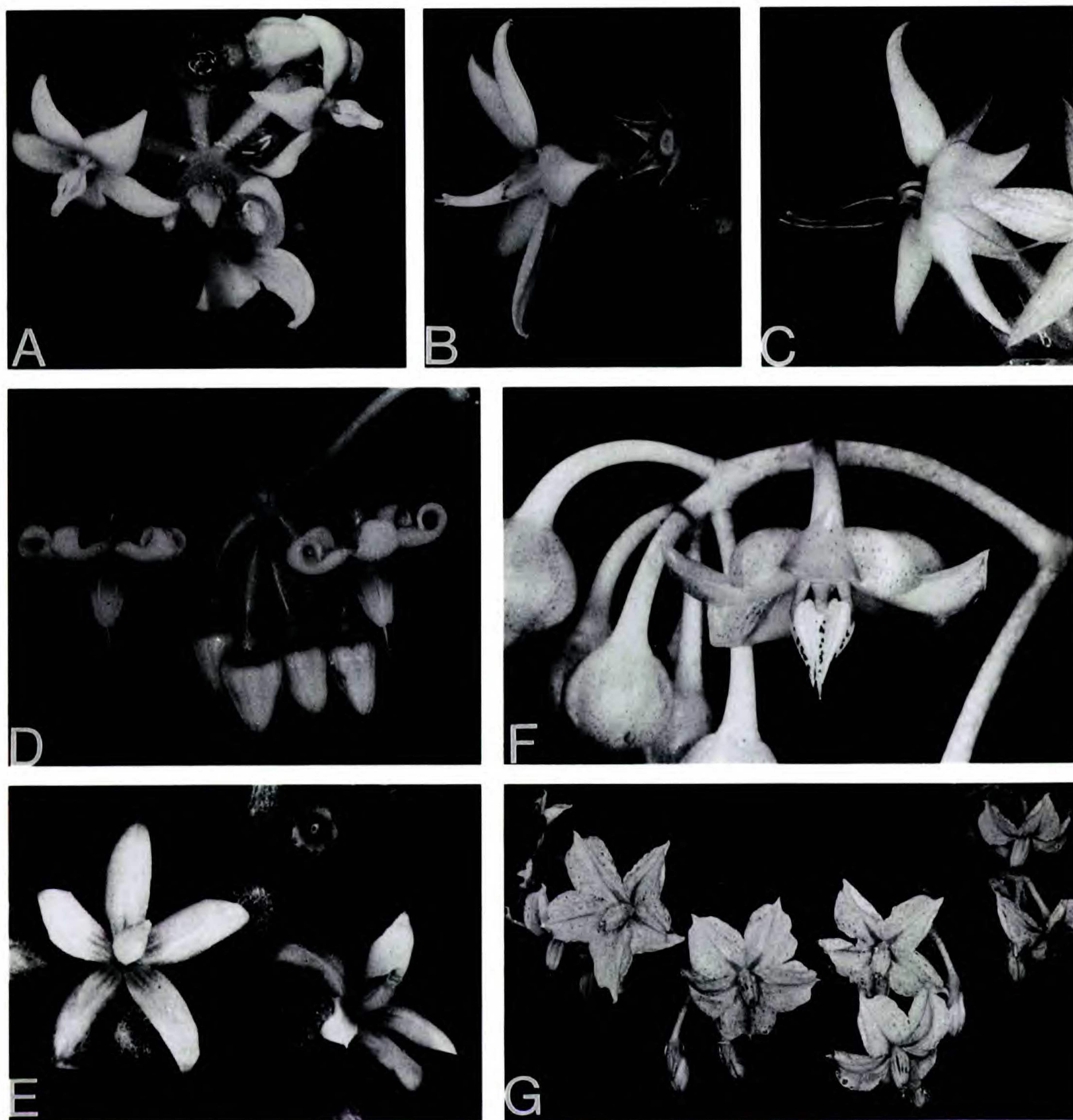
Figure 4.   A–D, Flowers of *Argostemma*.—A. *A. moultonii*.—B. *Argostemma* sp.—C. *A. diversifolium*.—D. *A. pictum*.—E. Flowers of *Strumpfia maritima*. F, G. Buzz-pollinated flowers of non-rubiaceous taxa.—F. *Ardisia* sp. (Myrsinaceae).—G. *Solanum* sp. (Solanaceae).

Also note the shape of the anthers in cross section (Fig. 3D): as compared to *A. borragineum* (Fig. 2E), the rounded connectives are rather small, and the abaxial pollen sacs of each anther are roughly in the same plane as the connective. Consequently, the anther tube is not a 5-*ridged* structure (ridges = connectives) as in *A. borragineum*, but more or less regularly 5-angled. Such a 5-angled anther tube is also found in *A. pictum* (Fig. 4D).

Species with poricidal dehiscence of the pollen sacs and (a) connivent anthers and (b) a more or less zygomorphic anther arrangement, as ex- emplified by A. diversifolium *and* A. neurocalyx, *respectively.*

The anthers of *Argostemma diversifolium* are held in a tubelike arrangement (Fig. 4C), but no fusions or agglutinations are present. The arrangement of the androecium often is slightly zygomorphic because all five filaments have an S-like curvature (thus moving the base of the anthers away from the center of the flower), and because the anthers themselves are also a little curved (Fig. 4C). Such slight curvatures also occur in *Argostemma* species with solid anther tubes, long, sterile
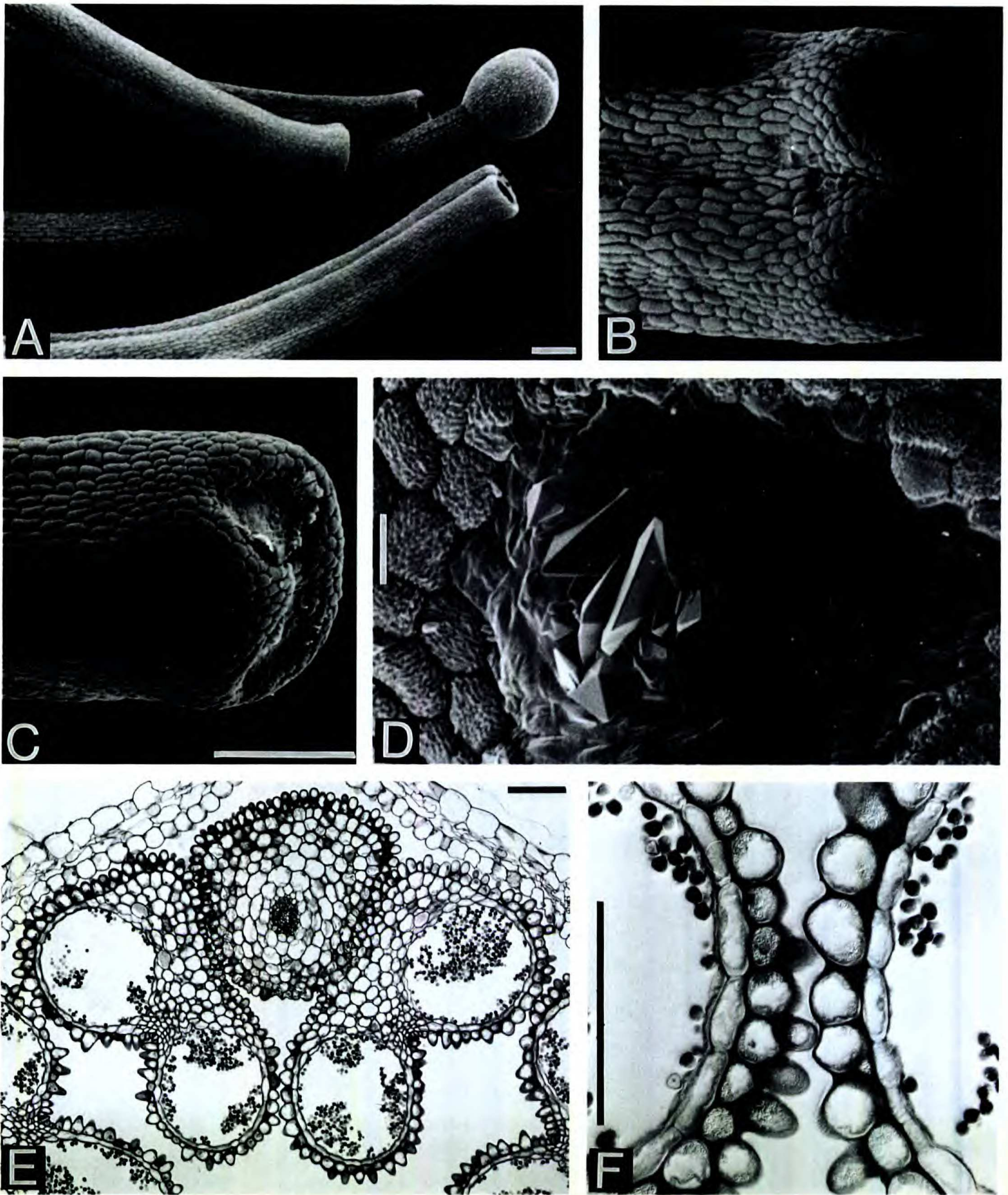
Figure 5.   *Argostemma diversifolium.* —A. Upper parts of stamens, style, and stigma (compare with Fig. 4C).— B. Poricidal dehiscence of anther (from open flower).—C. Apical part of anther (from bud) showing areas where the pores will be formed.—D. Detail of pore-forming area; note calcium oxalate crystals.—E. Cross section of anther near its base (bud shortly before anthesis, note corolla outside the anther).—F. Detail of pollen sacs of two adjacent anthers; note absence of thickenings in the endothecium. A–D: SEM-graphs; all others: LM-graphs. Scale bars: 0.1 mm (A; B = C; E; F); 10 μm (D).

apical appendages, and longitudinal dehiscence of the pollen sacs; the unidentified species from peninsular Thailand depicted in Figure 4B is a good example.

In this species, the anthers are prolonged for some distance below the insertion point of the filaments (so the anthers are not strictly basifixed as, for example, in *A. borragineum,* cf. Fig. 1B, or
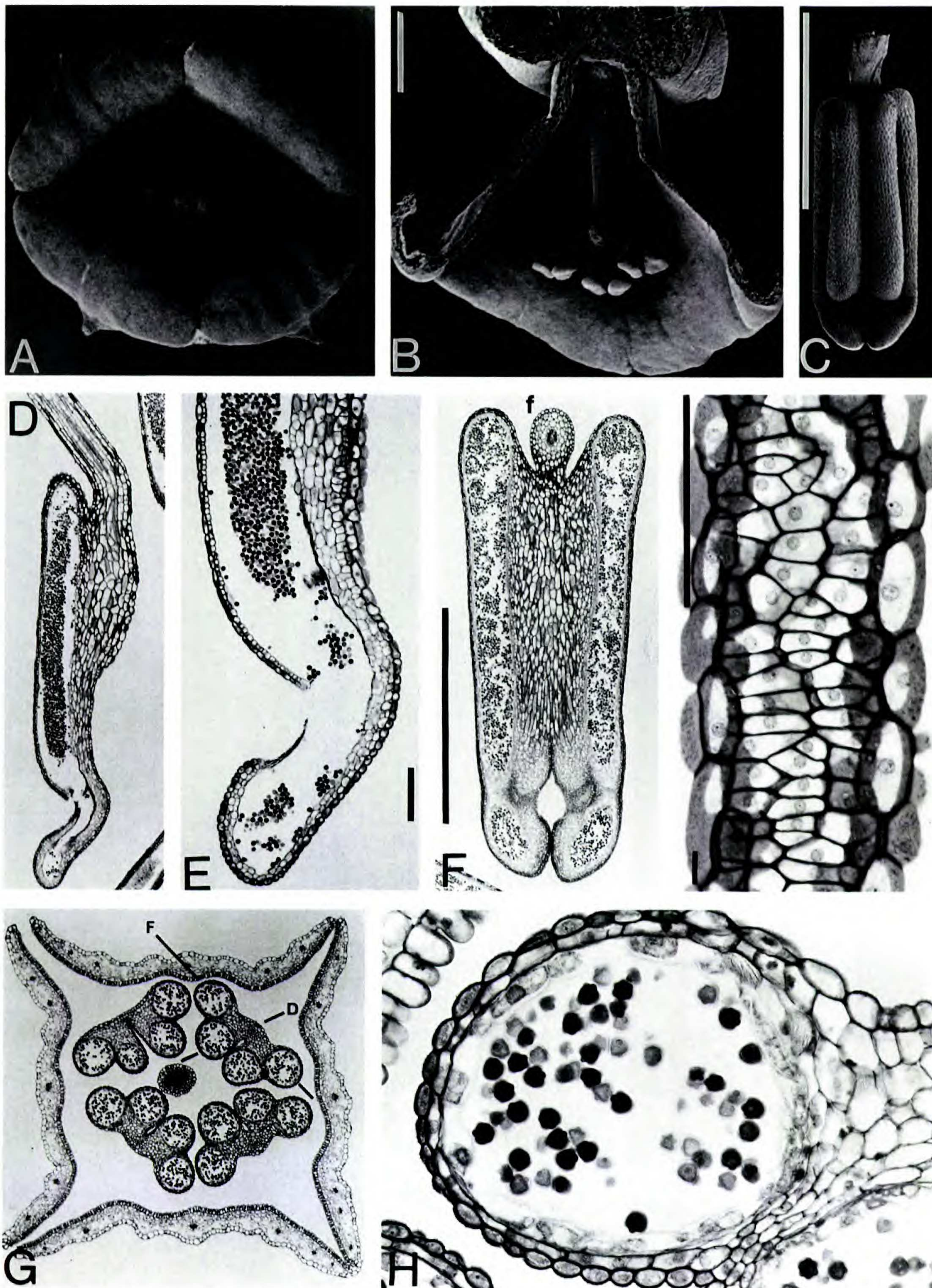
Figure 6.   *Argostemma neurocalyx.* — A. Front view of flower; note slight zygomorphy, enhanced by the position of the anthers (the fourth, slightly shorter stamen is not clearly visible). — B. Longitudinal section of flower, one stamen removed. — C. Isolated solitary stamen; note spoonlike structure formed apically by the two abaxial pollen sacs. — D. Off-median longitudinal section of stamen (approximate level of section indicated in G). — E. Detail of apical part of anther; note dehiscence pore. — F. Longitudinal section of stamen showing section of up-turned filament (f), connective, and abaxial pollen sacs (approximate level of section indicated in G). — G. Cross section of bud shortly before anthesis

A. moultonii, Fig. 4A) and are sagittate. Cross sections of the basal portions of the anthers thus show the free filament and a theca (= paired pollen sac) on either side of it (Fig. 5E). Above the insertion point of the filament, the connective is inconspicuous and occupies a relatively small area relative to the thecae. Apically, each anther has two round pores (Fig. 5A, B). In young anthers, the areas where the pores will be formed are easily recognizable as thin-walled slight depressions with large calcium oxalate crystals on their surfaces (Fig. 5C, D).

In contrast to the *Argostemma* taxa with longitudinally dehiscing pollen sacs, the pollen sac walls of *A. diversifolium* lack an endothecium with thickenings; there is merely a parenchymatic layer of "normal" cells below the pollen sac epidermis (Fig. 5F).

Argostemma neurocalyx represents yet another type of *Argostemma* flower. The 4-merous flower itself has a slightly zygomorphic, bell-shaped corolla (Fig. 6A). The arrangement of the anthers, all held near the two basal corolla lobes and in close proximity to each other, further enhances the zygomorphy (Fig. 6A, B). In nature, the flowers are held in a horizontal position.

There is no actual cellular fusion in the androecium. The abaxial and adaxial pollen sacs of each theca are of unequal lengths. The former are notably longer than the adaxial ones and form a kind of spoonlike structure (Fig. 6C) because their apical parts are slightly upturned (Fig. 6D, E). The abaxial side of the anthers is flat because abaxial pollen sacs and connectives are in one plane (cf. Fig. 6G). Whereas the abaxial pollen sacs are separated by the connective tissue (Fig. 6F, G), the adaxial ones are closer to each other (i.e., in a more central position), immediately above the connectives. These anthers each have two pores (i.e., each theca has one pore), which is common to one adaxial and one abaxial pollen sac. Because the abaxial pollen sacs are longer, this common pore is located some distance away from the anther apex, i.e., in the lowermost part of this spoonlike structure. Cross and tangential sections of the pollen sac wall (Fig. 6H, I) show the presence of a subepidermal, parenchymatic cell layer, but an endothecium, in the sense of a cell layer with fibrous thickenings, is absent.

Comments on anther dehiscence and endothecium.

The present investigations on *Argostemma* species suggest that there is a clear correlation between longitudinally dehiscent anthers and the presence of an endothecial layer with fibrous thickenings on the one hand, and the absence of thickenings in the subepidermal cell layer of the pollen sac wall in taxa with poricidal anther dehiscence on the other hand.

The suppression of the endothecium appears to be the rule in plants whose anthers open by pores (Goebel, 1923), and this phenomenon occurs both in monocots and dicots, notwithstanding their familial position and relationships (Ericaceae, Solanaceae, etc.). *Argostemma*, however, needs to be added to the relatively few examples where both situations occur in the same genus.

Comments on the pollination ecology.

Argostemma flowers belong to the "*Solanum*-type" of pollen flowers sensu Vogel (1978) (Fig. 4G). This "*Solanum*-type" is widespread and occurs in oligandrous families and orders, and in both monocots and dicots (including *Ardisia* spp., Myrsinaceae; Fig. 4F). Characteristics of the "*Solanum*-type" are, according to Vogel (1978), that the few, mostly (sub)sessile anthers are enlarged, thus becoming more showy and capable of producing excess pollen. The powdery pollen is released in small portions as a "cloud" by vibrating movements of the visitors, thus dusting their bodies. These "*Solanum*-type" flowers are strictly melittophilous and often (but not always) zygomorphic (compare the actino- and zygomorphic *Argostemma* species depicted in Fig. 4A–D), and the visitor clasps the androecium in a fixed position. Because of the overall dusting of the visitor, the position of the small stigma is independent from that of the anthers. In view of the widespread occurrence of floral nectaries in the Rubiaceae (normally produced on the "roof" of the inferior ovary), the absence of nectaries in *Argostemma* is noteworthy and congruent with the "*Solanum*-type" of pollen flowers.

As noted above, vibratile ("buzz") pollination is characteristic, and Buchmann (1978) does in fact include *Argostemma* in his table of buzz-pollinated

←

(note corolla lobes); sections of the upper half of the anthers.—H. Detail of pollen sac; the cell layer below the pollen sac epidermis has no thickenings.—I. Tangential section of endothecium; note absence of the thickenings. A–C: SEM-graphs; all others: LM-graphs. Scale bars: 1 mm (A = B; C; D = F = G); 0.1 mm (E; H = I).

angiosperms. Buchmann (1978, 1983), however, overemphasized the intimate link between buzz pollination and poricidally dehiscent anthers, and Vogel (1978) also mentioned only poricidal anthers as being a characteristic of "*Solanum*-type" flowers.

We believe that those *Argostemma* species with longitudinally dehiscent anthers and an anther tube are also buzz pollinated. The formation of an apically constricted tube leads to the same effect as apical pores because there is always a terminal opening; the exserted style never entirely blocks this tube but always leaves enough room for pollen to exit. Support for this interpretation comes from Sazima et al. (1993: 80), who noted, "It must also be kept in mind that flowers with longitudinal slits may form, using other morphological elements, tubular structures analogous to the buzz anthers." In his study of *Lycopersicon* (Solanaceae), a genus which, like many *Argostemma* species, has solid anther tubes with sterile apical appendages and longitudinally dehiscent anthers, Teppner (1993) provided actual photographic proof of buzz pollination by bees.

A (far from complete) survey of *Argostemma* species shows that the presumably buzz-pollinated species show great diversity in shape, size, color, and arrangement of the androecium and also in the shape and outline of the (most probably always) white corolla.

In some species (e.g., *Argostemma moultonii*, Fig. 4A), all floral parts are pure white, or in others (e.g., *A. chaii*, *A. neurocalyx*, Fig. 6A) white except for the yellowish or yellow to pale orange stamens (both filaments and anthers). In *A. pictum* (Fig. 4D), the anthers are yellowish above, but basally the connectives and short filaments are green. Not uncommon are green spots near the base of the corolla lobes in addition to the green filaments, yellow anthers, and whitish apical connective appendages (e.g., *A. yappii*; Bremer, 1984: fig. 1C: color illustration). *Argostemma diversifolium* (Fig. 4C) may be the only species with blue anthers, green filaments, and green spots near the insertion points of the filaments. In the *Argostemma* species depicted in Figure 4B, the swollen, somewhat enlarged filaments are orange (dark in the b/w reproduction), and anthers as well as apical connective appendages are very pale yellow to whitish.

In all but one of the examples given above, the corolla shapes range from subrotate to subcampanulate, and the lanceolate corolla lobes are spreading to recurved or reflexed. *Argostemma neurocalyx* has a small (ca. 5 mm long) campan-

ulate, slightly compressed (and, therefore, slightly zygomorphic) 4-merous corolla with short recurved lobes, and the entire appearance of the flower is reminiscent of that of certain Gesneriaceae, such as small-flowered *Didymocarpus* species, also because all four anthers are located in the vicinity of the "lower lip."

STEENISIA (FIG. 7)

Existing information on the androecium of this small genus (5 species, all occurring in Borneo) is superficial. Bremer (1984) described its condition only as "anthers adnate to an ovate cone; thecae with two loculi; connective produced apically above the thecae" (p. 337) and "thecae open by vertical slits" (p. 334); the relevant illustrations are too imprecise to draw any definite conclusions.

The species studied, *S. pleurocarpa*, has a subsessile, pale yellow anther tube (the free filaments < 0.5 mm long) which is ca. 4–5 mm long. Only the lower two-thirds or less of the entire tube is occupied by the pollen sacs, whereas the "apical" remainder is exclusively formed by flattened connective appendages (Fig. 7B, D). Each of these appendages is, on average, 5–6 cells thick and contains a weakly developed vascular bundle in the center (Fig. 7E). The most apical parts of these appendages are rounded and free from each other. Slightly below the apex, however, the connective appendages form a solid, not easily ruptured, angled tube or hollow cone. The neighboring appendages are interlocked in a peculiar fashion (Fig. 7F); they are fused by their cuticles. Similarly, the epidermis cells of pollen sacs of neighboring anthers are united by fusion of their cuticles in the lower half of the anther tube (Fig. 7G, H). Consequently, the entire anther tube, from bottom to top, is a solid unit.

Upon maturity, the pollen is released into the interior of this tube. The pollen sacs dehisce longitudinally. The line, where the pollen sacs will dehisce, is already clearly visible in late bud stage (Fig. 7H, triangle). The endothecium of the anthers has ringlike thickenings (visible in some cells, but not yet fully developed in the anther section depicted in Fig. 7H; from a bud shortly before anthesis; note that tapetum remnants are still present).

According to Bremer's (1984) descriptions and illustrations, *S. pleurocarpa* has 5-merous flowers and white corollas. However, the study material and other populations of this species seen in Borneo all had only 4-merous flowers (cf. Fig. 7G) and pink corollas.
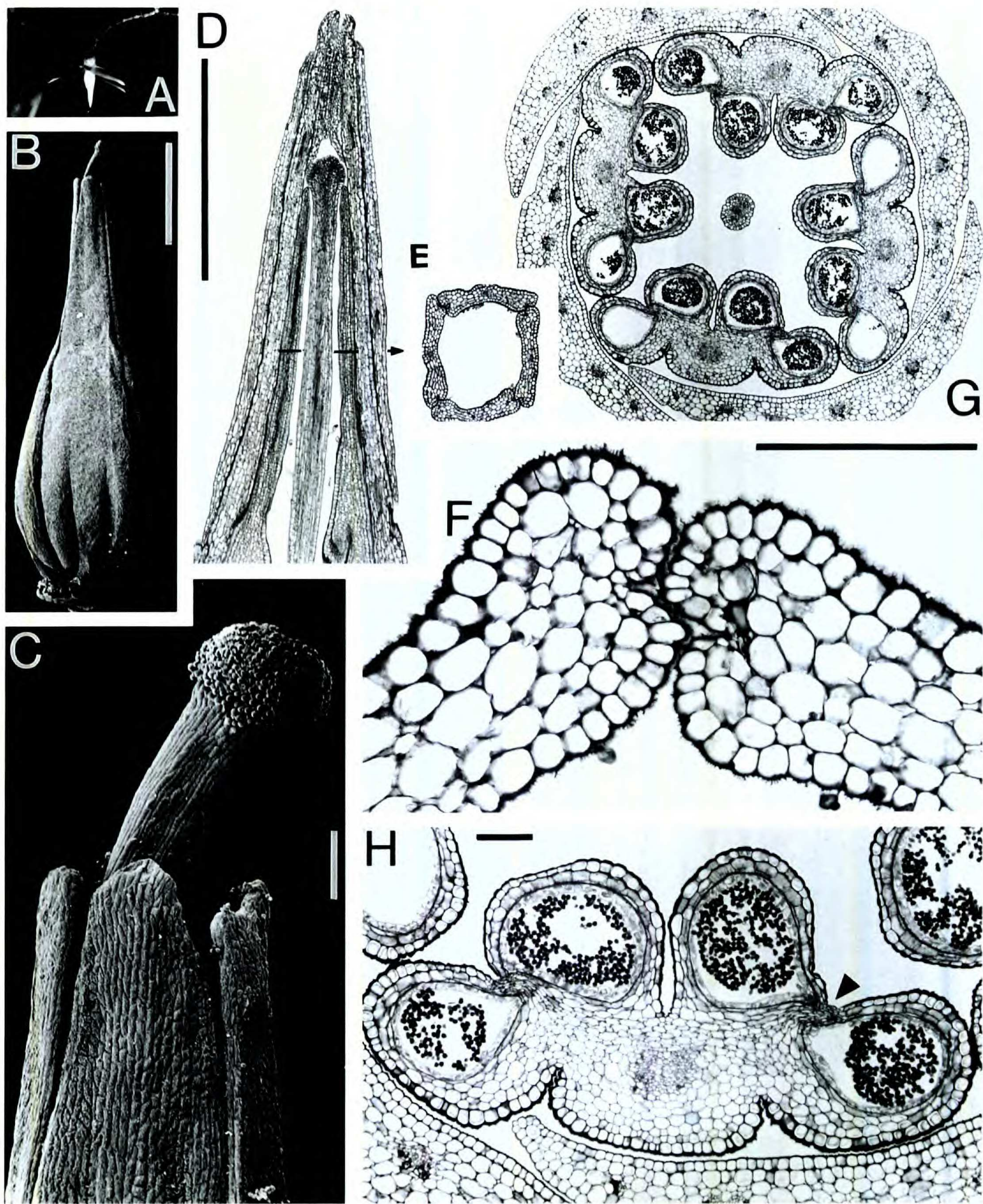
Figure 7. *Steenisia pleurocarpa.* —A. Open flower in natural, pendulous position (corolla pink; anther cone pale yellow, style and stigmas white).—B. Anther cone of flower at anthesis, corolla removed.—C. Detail of A, showing free ends of apical connective appendages, and the exserted, always slightly curved style and stigma.—D. Longitudinal section of uppermost part of bud shortly before anthesis; visible from out- to inside: corolla, connective appendages and uppermost part of anthers (below), style, and stigma.—E. Cross section of connective appendages at the level indicated in D.—F. Detail of E, interlocked connective appendages of two neighboring stamens.—G. Cross section through the lower third of a flower bud shortly before anthesis (from out- to inside: corolla lobes, anthers, style).— H. Detail of G; the lateral pollen sacs of the anther in the center are in close contact but not fused with those of the neighboring anthers (triangle: area of pollen sac dehiscence). A and B: SEM-graphs; all others: LM-graphs. Scale bars: 1 mm (B; D = E = G); 0.1 mm (C; F; H).

Comments on the pollination ecology.

There can be no doubt that the flowers of all *Steenisia* species, like those of *Argostemma*, belong to the buzz-pollinated "*Solanum*-type" of pollen flowers sensu Vogel (1978). Variation in corolla shape and size and androecium color and arrangement is not as great as in *Argostemma*, and the corollas are generally star-shaped, ranging only in color from white to pink. The entire androecium (anther tube plus filaments) is pale yellow to orange (cf. Bremer, 1984: fig. 1A, color illustration of *S. borneensis*). As in *Argostemma*, a nectar-producing disk is absent.

In all but one *Steenisia* species the styles are beset with upwardly directed hairs (cf. relevant figures in Bremer, 1984). These hairs do not extend to the uppermost part of the style but are generally confined to the region that is at the same level as the pollen sacs. It seems plausible to conclude that their function is to "catch" released pollen.

It should also be noted that in all *Steenisia* species the upper part of the style invariably is curved (Fig. 7A, C; also cf. relevant illustrations in Bremer, 1984). The pollination biological relevance of this feature may not be great; compare in this context the information given on general characteristics of "*Solanum*-type" of pollen flowers under *Argostemma*.

STRUMPFIA (FIG. 4E)

This monotypic Caribbean genus stands out among all other Rubiaceae with fusions or agglutinations in the androecium in that it is the only known taxon in which all five anthers are united into a tube by means of a discrete cell layer (a kind of "super epidermis"). Refer to Igersheim (1993a) for illustrations and detailed information on additional peculiarities such as the formation of partially united, crownlike apical connective appendages, the retraction of the style in the functionally male stage, and the proterogyny in this genus.

Presumably on the basis of literature data, Buchmann (1978) had included the genus in his list of buzz-pollinated taxa. One of us (Igersheim, in prep.) is now able to provide proof and photographic documentation for the occurrence of buzz pollination.

ACRANTHERA (FIGS. 8–11)

Acranthera is an Asiatic genus of ca. 35 species, whose taxonomic position to this day is not entirely resolved. The reason for this unsatisfactory situation is the lack of detailed data on most of its character states. In floral characteristics, the genus is rather diverse, as reflected by Bremekamp's (1947) infrageneric subdivision into seven subgenera, whereby some of these are further split into "series."

The present investigation is restricted to three of Bremekamp's subgenera: (1) Subgenus *Phanerochiton* Bremek. (*A. involucrata*) and (2) subgenus *Dichroanthes* Bremek. (*Acranthera* sp., aff. *A. variegata*), both of which are distinguished by an elongated "receptaculum pollinis" sensu Bremekamp; the two are primarily separated by inflorescence characters. (3) Subgenus *Mitracme* Bremek., characterized by a mitriform "receptaculum pollinis" sensu Bremekamp that is much shorter than the thecae; the four "series" he recognized within the subgenus are distinguished by "eciliolate" or "ciliolate connectives" (e.g., series Ciliolatae: *A. frutescens*), and by inflorescence shape (umbel-like inflorescences: series Umbelliflorae: *A. atropella*).

The genus *Acranthera* is characterized by the presence of united apical connective appendages. Invariably, only the very apical parts of these appendages are fused (e.g., Figs. 8H, 10E). Microtome sections (Fig. 9A, B) reveal that this unification comes about by epidermal cells of neighboring connective appendages being interlocked, coupled by the fusion of their cuticles.

The length of the sterile, apical connective appendages varies from short (e.g., *A. involucrata*, Fig. 8E, G) to relatively long (e.g., *A. frutescens*, Fig. 10C). Appendage length is obviously correlated with the size and shape of the stigmatic areas (see below for details).

The (typically) 5, elongated and rather narrow anthers have dorsally prominent connectives that are either rounded (cf. Figs. 8F, 10A, B, 11C) or pointed (triangular in cross section, Fig. 9F, G). Viewed from above, the anther complex is a 5-lobed structure (Figs. 8A, 10A, 11C).

In the investigated species, the anthers either have hairs only in the border area between connective and pollen sacs (i.e., a line of hairs laterally on either side of the connective; *A. frutescens*, Fig. 10B, D), or also on the pollen sacs (*A. involucrata*, Fig. 8B, F, G). Bremekamp's (1940) distinguishing character, "ciliolate connectives," therefore, is rather imprecise.

Not only in the studied, but presumably in all taxa of the genus, the pollen sacs open with longitudinal slits (Figs. 8G, 10C, D). Pollen is thus released into the interior of the anther complex.

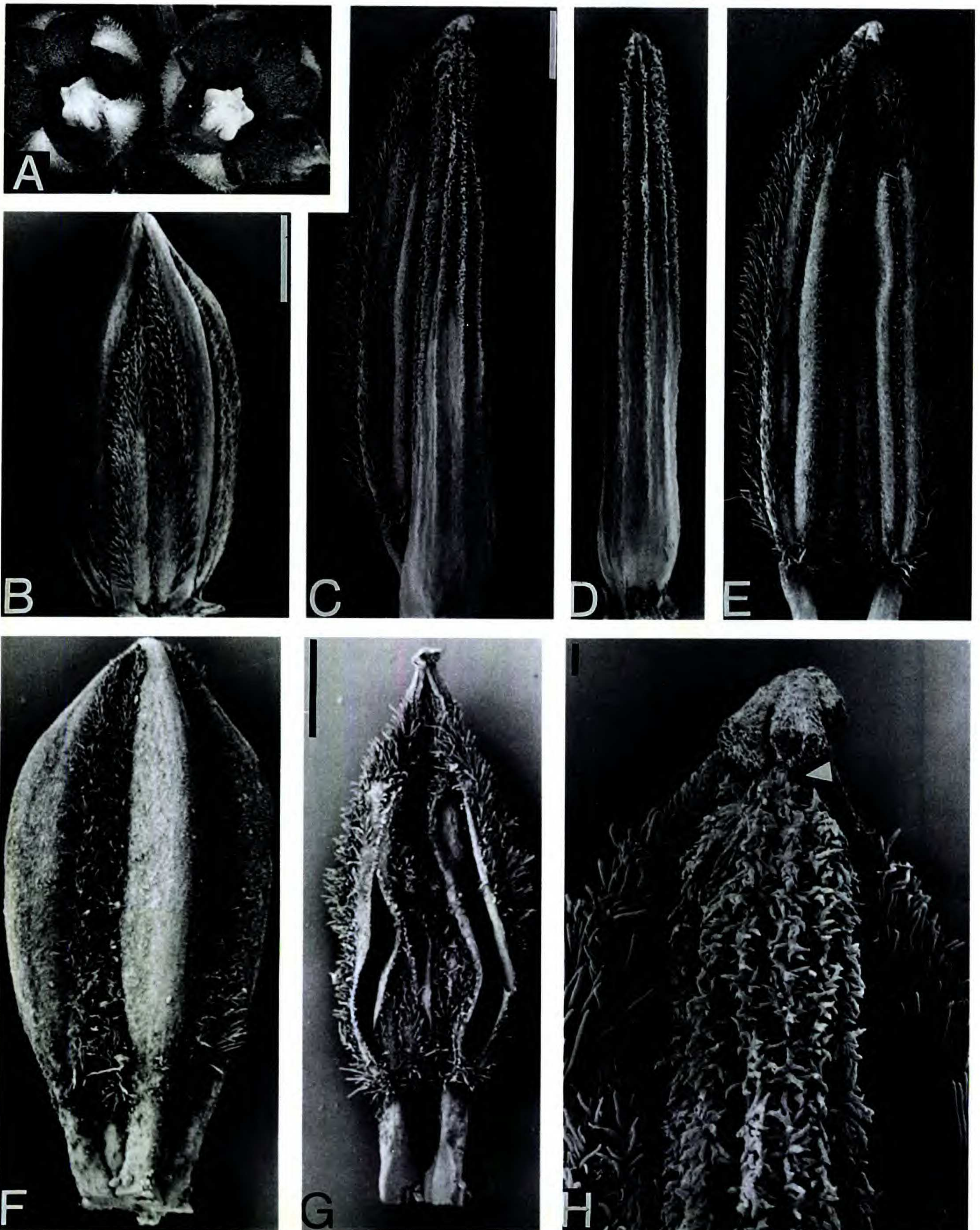All taxa covered by the present study have very

Figure 8.   *Acranthera involucrata* (subg. *Phanerochiton*).—A. Pinkish, short-tubed flowers with exposed, white anther complexes.—B. Anther-style and stigma complex from young bud; note apical connective appendages. C–E. From bud older than in B.—C. Anther-style and stigma complex, several anthers removed to show style and stigma.— D. Isolated style and stigma (anthers removed). Receptive areas not yet fully developed; compare with H.—E. Two isolated stamens held together by apical connective appendages; style and stigma removed. Note hairs on the pollen sacs. F–H. From open flower.—F. As B, but note "hairy windows" between anthers.—G. As E, but pollen sacs dehisced.—H. Detail of the apical region; note fusion between apical connective appendages and the stigma (triangle), and the receptive surface of the stigma. A–H: SEM-graphs. Scale bars: 1 mm (B; F = G; C = D = E); 0.1 mm (H).
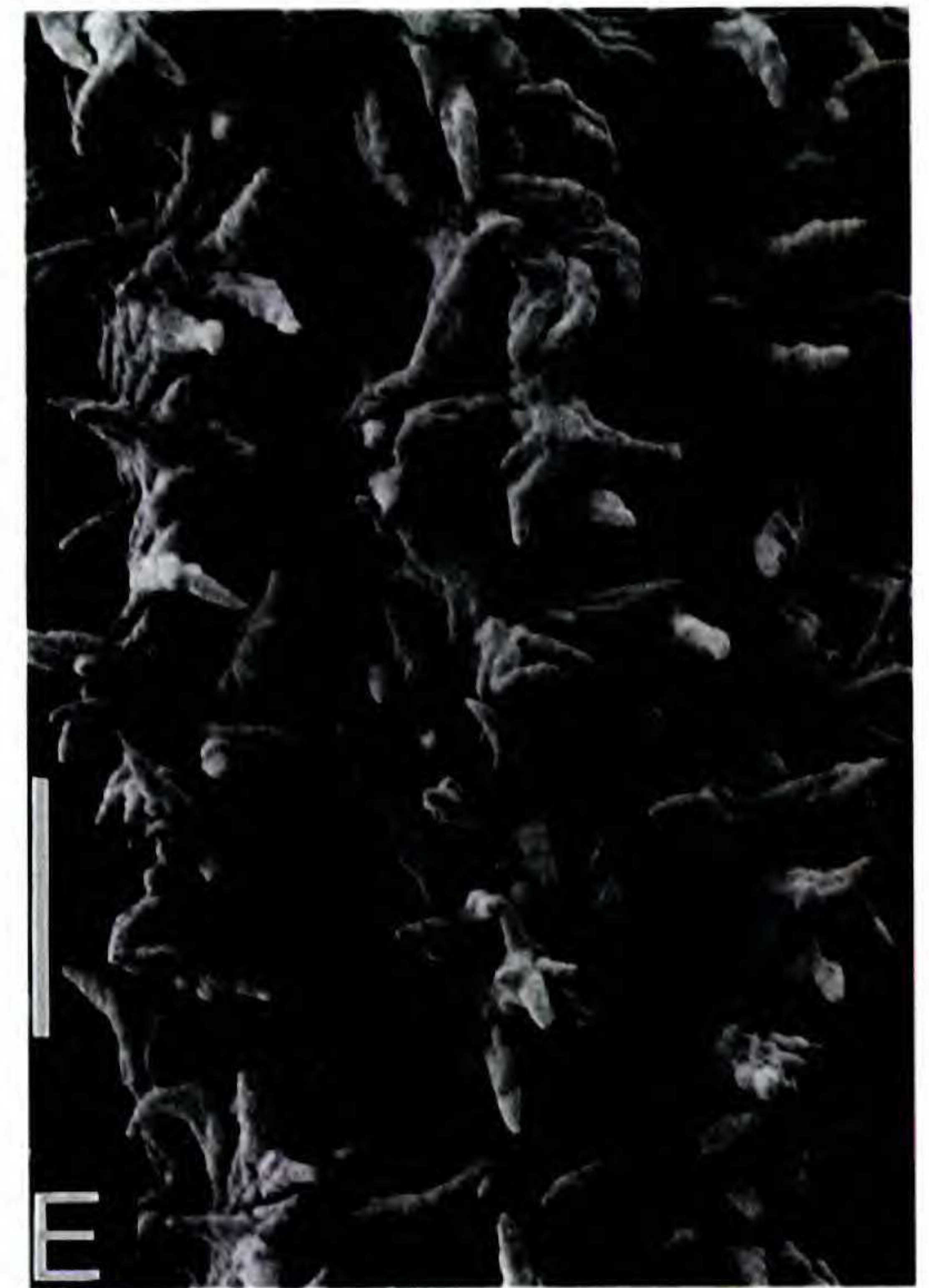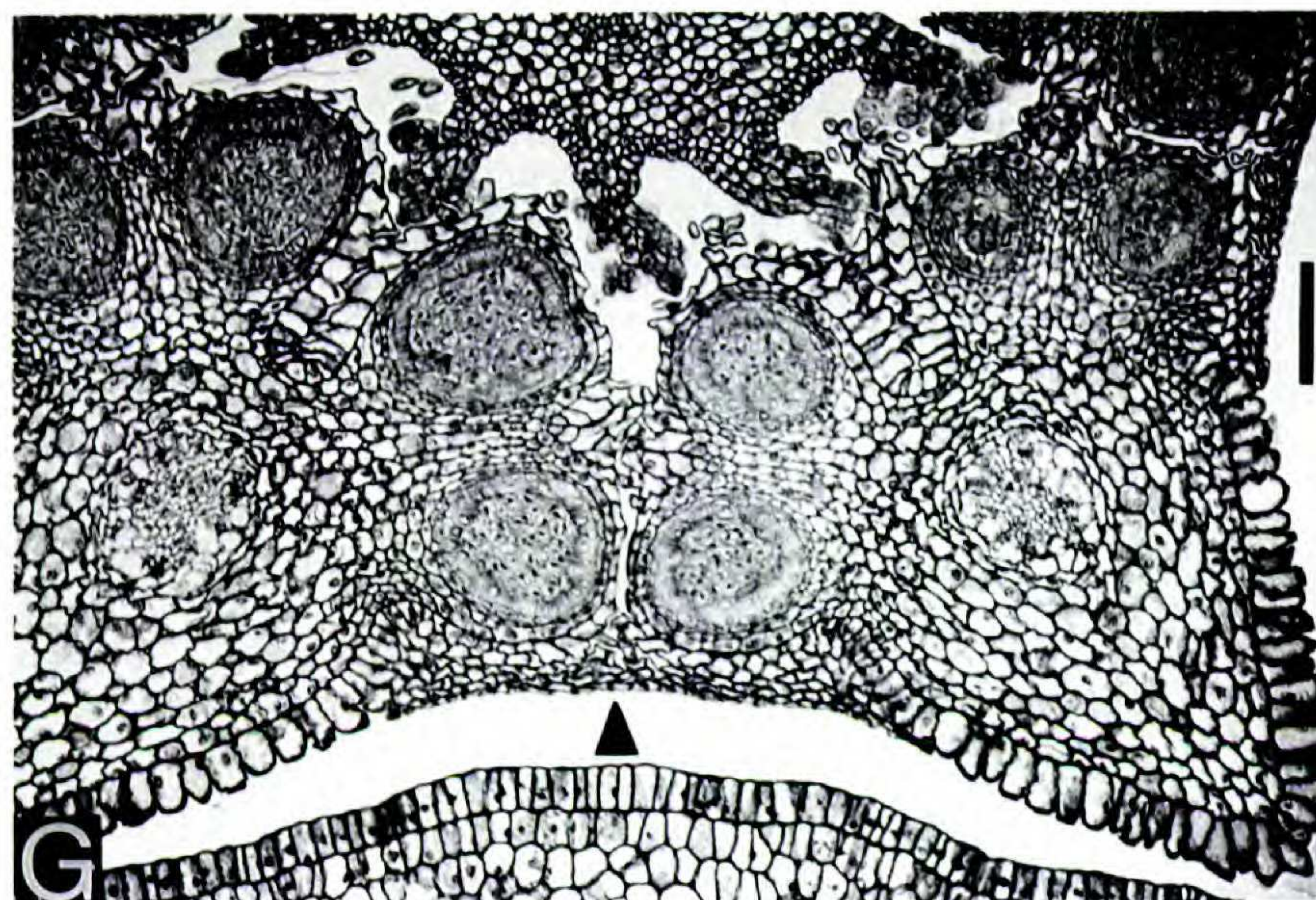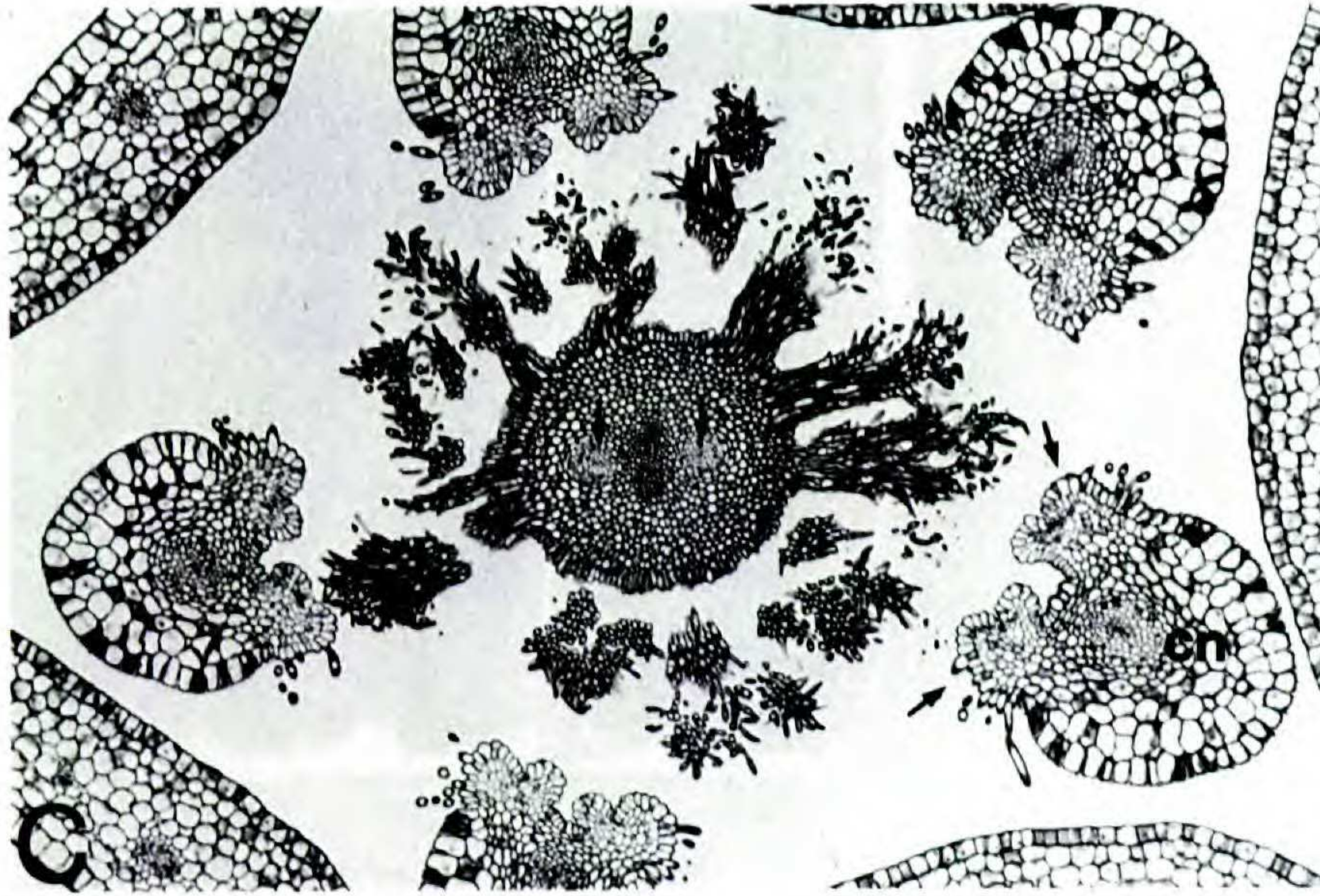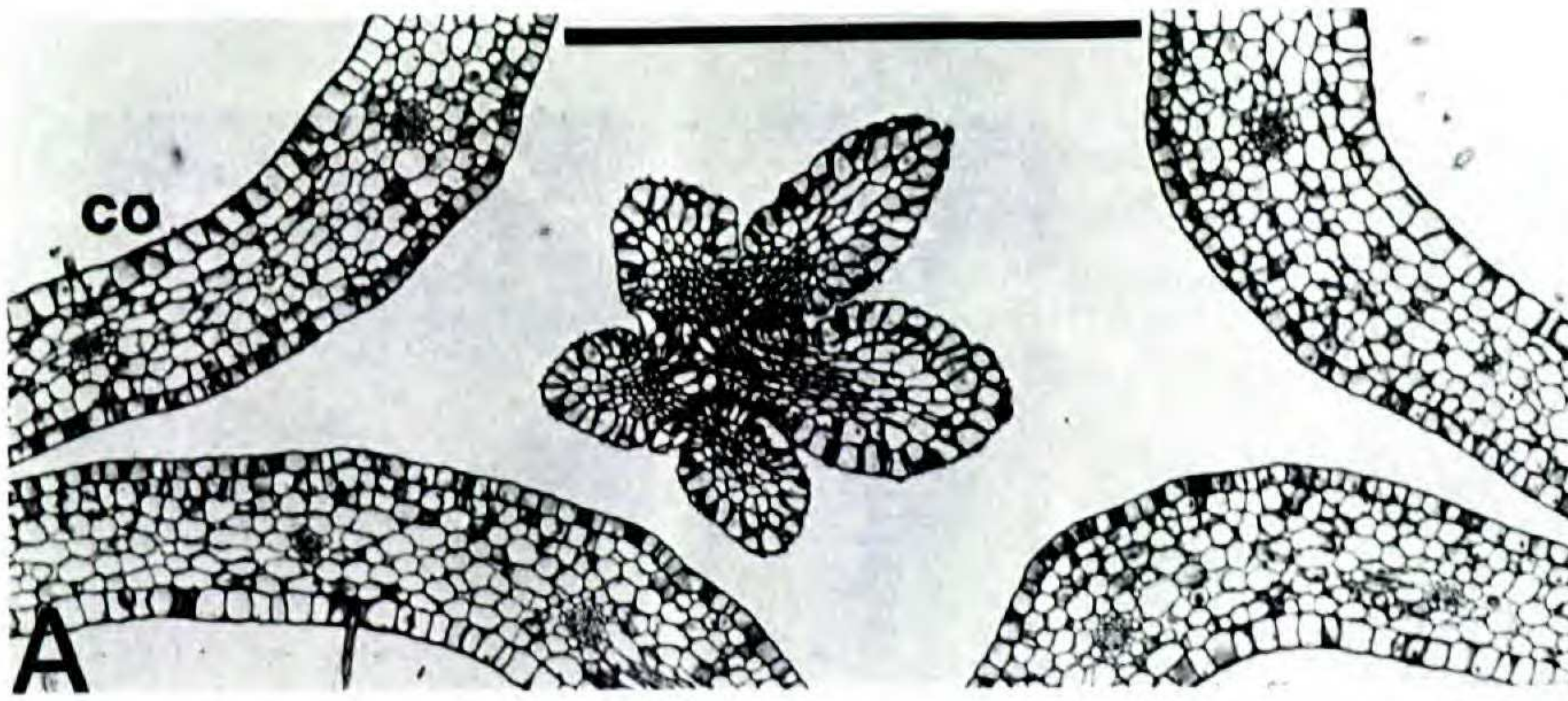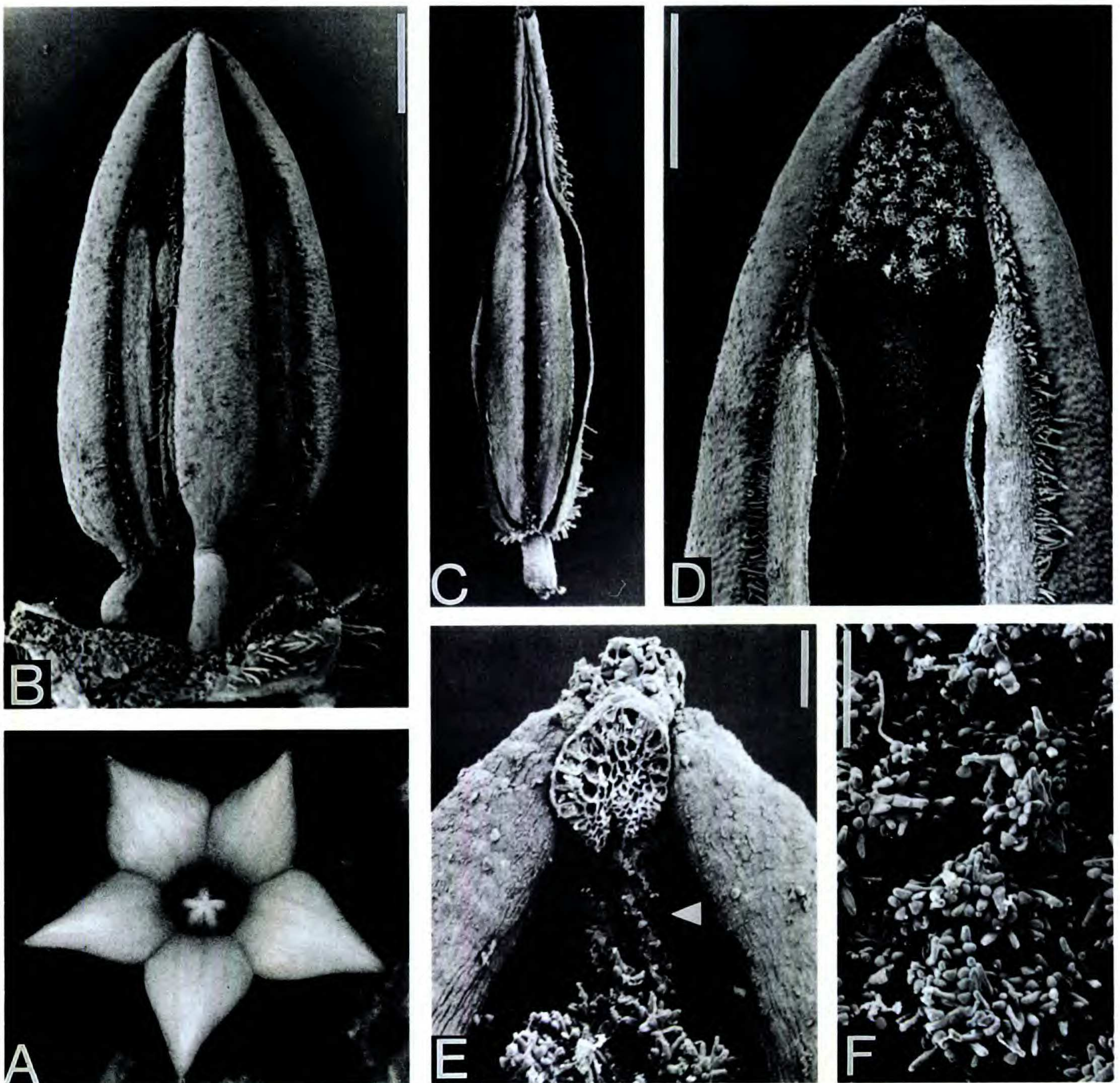
Figure 10.   *Acranthera frutescens* (subg. *Mitracme*, series *Ciliolatae*).—A. Greenish yellow, ± bell-shaped flower from above; the whitish anther complex is included in the tubular part of the corolla.—B. Anther-style and stigma complex from open flower; note "windows" above pollen sacs.—C. Isolated anther; note dehisced pollen sacs and long apical connective appendage.—D. Upper part of anther-style and stigma complex, one stamen removed to show mitriform stigma.—E. Columnar tissue (triangle) connecting stigma with connective appendages.—F. Detail of stigmatic area (receptive surfaces). A–F: SEM-graphs. Scale bars: 1 mm (B = C; D); 0.1 mm (E; F).

←

Figure 9.   A–E. *Acranthera involucrata* (subg. *Phanerochiton*). A–D. Cross sections of bud shortly before anthesis.—A. Uppermost part, united connective appendages (co, corolla lobe).—B. Detail of A, showing tissue fusion of connective appendages.—C. Slightly lower level than A, showing connectives (cn) with sterile, uppermost parts of the thecae (arrows) and fingerlike, branched stigmatic areas (arrows in the central column of the stigma point to loose cells, which most probably correspond to the region where the receptive stigmatic papillae or hairs of a "typical" bicarpellate Rubiaceae with two stigma lobes would be located).—D. Detail of "stigma-fingers" with branched endings.—E. Rows of branched stigmatic papillae. F, G. *Acranthera* sp., aff. *A. variegata* (subg. *Dichroanthes*), cross sections of bud shortly before anthesis.—F. Corolla, anthers and stigmatic "fingers."—G. Detail of F, two neighboring anthers still connected by fragile, parenchymatic tissue (triangle; this will tear at anthesis, resulting in the formation of longitudinal slits between anthers). E: SEM-graph; all others: LM-graphs. Scale bars: 1 mm (A = C = F); 0.1 mm (B = D; E; G).
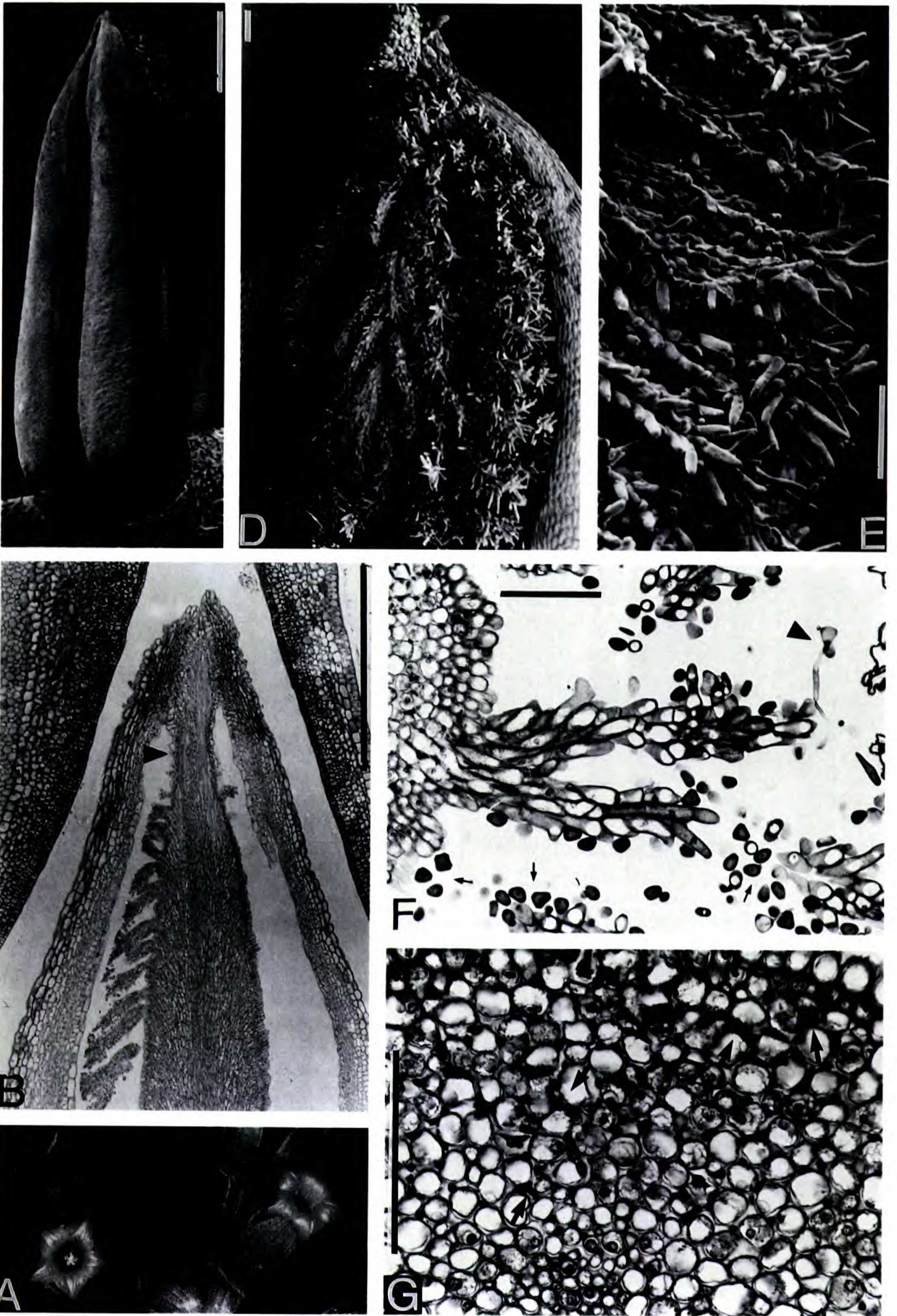
short (Figs. 8F, G, 10B) or subobsolete filaments (Fig. 11C). This is not uniformly so in the genus; see comments below.

A unique feature of *Acranthera* is that the apically united connective appendages are, in turn, united with the stigma by means of a columnar tissue (Figs. 8H, 10E, 11B, D). The latter is a solid, parenchymatic tissue, which is an upward extension of the central portion of the stigmatic tissue (Fig. 11B). This intimate connection between connective appendages and stigma was already present in the youngest floral developmental stages available for investigation (i.e., seemingly very young buds), indicating that this fusion, although undoubtedly postgenital in nature, must come about at very early stages of development.

The presence of this "anther-style and stigma complex" prompted Bremekamp (1947: 237) to state "[This] is a feature so entirely unparalleled in the Rubiaceae, that one may feel inclined to regard the position of the genus with regard to the rest of the family, as similar to that of the Asclepiadaceae with regard to the Apocynaceae, although in this way the importance of the connection between the anthers and the style is doubtless overemphasized."

The major misconception in Bremekamp's (1947) monograph of *Acranthera* is that he continually emphasized the presence of a "receptaculum pollinis." He stressed (p. 271) that "the pollen is temporarily deposited on the upper part of the style, which, to that end, is covered by rows of papillae."

The detailed studies discussed below prove that the situation is entirely different.

In *Acranthera involucrata*, style and stigma form a 10-ridged, uniform columnar structure (Fig. 8C, D; from a young bud). At this stage of anthesis no part of the style or stigma is visible from outside. The anthers bear relatively short apical connective appendages so that, in contrast to other investigated species, the anther complexes of open flowers do not show conspicuous apical "windows" (= gaps between connective appendages; Fig. 8F, compare with Figs. 10B and 11C).

The 10 ridges of the style and stigma column "fit" perfectly into the gaps between pollen sacs of neighboring anthers, and in the depressions between the two adaxial pollen sacs of each anther (cf. *Acranthera* sp., aff. *A. variegata*, Fig. 9F).

In bud stage, the upper half of the style and stigma column bears indistinct (not yet fully developed) hairlike appendages (Fig. 8C, D) on the ridges. During further development (from buds shortly before anthesis onward), the ridges grow out into fingerlike structures, and their irregularly arranged, hairlike appendages, whose ultimate parts are uniseriate and uni- or pluricellular, become very prominent (Fig. 9C, D). Figures 8H and 9E show such rows of irregularly arranged, hairlike appendages. Cross sections from the upper third of this style and stigma column reveal that the "central core" always shows two vascular bundles and, at a right angle to them, two areas (or sometimes only a single, uniform area) of looser cells (Fig. 9C; also cf. *Acranthera* sp., aff. *A. variegata*, Fig. 9F).

The interpretation of the hairlike appendages borne on the fingerlike structures described above is that they must represent the actual receptive (stigmatic) areas. Two stigma lobes as such (bicarpellate gynoecium) are never developed. The loose cells perpendicular to the two vascular bundles (see above) most probably correspond to the region where the receptive stigmatic papillae or hairs of a "typical" bicarpellate Rubiaceae with two stigma lobes would be located (cf., for example, *Crocyllis*, Puff & Mantell, 1982: fig. 15).

Acranthera is thus unique in the Rubiaceae in having an "anther-style and stigma complex" and also in the structure of its stigmas.

In the Rubiaceae a "receptaculum pollinis," or pollen-presenting organ, is typically the nonreceptive style, or style plus the nonreceptive outside of the actual stigmas, both of which may be provided with special adaptations (hairs, sticky substance, enlarged, specialized structures; cf. Igersheim, 1993b: fig. 1). Moreover, this mechanism of secondary pollen presentation is most probably always linked with protandry: the stigma lobes only unfold

---

Figure 11. *Acranthera atropella* (subg. *Mitracme*, series *Umbelliflorae*).—A. Pale yellow, ± bell-shaped flowers; the whitish anther complexes are included in the tubular parts of the corollas.—B. Longitudinal section of uppermost part of bud shortly before anthesis; visible from out- to inside: corolla, connective appendages, stigmatic area, and columnar tissue connecting apical connective appendages and stigma (triangle). C–G. Open flower.—C. Corolla removed to show anther-style and stigma complex; note stigmatic areas protruding from windows above pollen sacs.—D. Stamen removed to show mitriform stigma.—E. Stigma "fingers" with receptive hairs.—F. Cross section of stigma "finger"; note pollen grains (small arrows) and germinating pollen grain (triangle).—G. Cross section of the central column of the stigma with numerous pollen tubes (arrows). B, F, and G: LM-graphs; C–E: SEM-graphs. Scale bars: 1 mm (B; C); 0.1 mm (D; E; F; G).

and become receptive after the pollen has been shed (and carried away by pollinators). In view of this, *Acranthera*'s "receptaculum pollinis" sensu Bremekamp (1947) is not comparable to that of other Rubiaceae.

While in the *Acranthera* species described above, the unusual stigmatic areas may, to some extent, also function to "catch" pollen from its own anthers, their main and primary function is to receive "foreign" pollen. At this stage, there is, unfortunately, no information on whether the species is self-compatible or self-incompatible.

In *Acranthera frutescens* and *Acranthera atropella*, it is more obvious that the stigmatic (receptive) areas have nothing to do with secondary pollen presentation, and definitely do not function as a "receptaculum pollinis." In both species, the apical connective appendages are relatively long, and the gaps between the individual appendages form five "windows" above the pollen sacs (Figs. 10B, 11C).

During later stages of anthesis (i.e., after the pollen sacs have dehisced), the stigmatic areas protrude from these windows (Fig. 11C). As compared to *A. involucrata* (and all species of subg. *Phanerochiton* and *Dichroanthes*), the stigmatic areas are short and mitriform, their lengths corresponding to the lengths of the apical "windows" (Figs. 10D, 11D). As in the previously described species, there is no indication of the presence of two "normal" stigma lobes.

The stigmatic "fingers," moreover, differ in size and fine structure. In both *A. frutescens* and *A. atropella*, they are considerably longer than in *A. involucrata*. *Acranthera frutescens* has shorter stigmatic "fingers" than *A. atropella*, and each "finger" bears numerous, short hairlike appendages that are arranged over the entire surface (Fig. 10F). In *A. atropella*, the elongated, obliquely downward-directed "fingers" (Fig. 11D, E) are sometimes forked near their base (Fig. 11F). Terminally, the "fingers" often bear pluricellular, uniseriate hairlike appendages, which frequently are in an irregularly starlike arrangement.

In *Acranthera frutescens* and *A. atropella*, stigmatic areas and the region of the pollen sacs are clearly spatially separated. It is therefore not possible that the stigmatic areas can serve as a "receptaculum pollinis" for pollen from its own anthers. When the pollen is shed, it must drop to the bottom, i.e., on to the roof of the inferior ovary. From the viewpoint of pollination ecology (see also below) it is relevant to stress that in all *Acranthera* species investigated by us a nectar-producing disk (in Rubiaceae normally developed on the roof of the ovary) was absent.

In *A. atropella*, we frequently detected in SEM-graphs and microtome sections that numerous pollen grains are caught in between the stigmatic areas. Sometimes, grains even showed pollen tubes (Fig. 11F). A detailed investigation of the central, columnar core of the stigmatic area often revealed the presence of numerous pollen tubes in the intercellular spaces of the parenchymatic tissue (Fig. 11G). We believe that these flowers had been visited by pollinators, and that the pollen and pollen tubes present are from another flower. We do not have proof of the presence of a self-incompatibility system, however.

Comments on the pollination ecology.

In spite of the fact that actual field observations are lacking, several comments on pollination ecological aspects seem appropriate.

It is believed that the investigated *Acranthera* species, especially those of subgenus *Mitracme* (*A. frutescens* and *A. atropella*), are buzz pollinated.

The size and shape of the nectarless flowers, as well as of the "anther-style and stigma complex" (< 10 mm long), agree well with "typical" oligandrous buzz-pollinated flowers. The five apical windows (*A. frutescens* and *A. atropella*) could be likened to apical pores of buzz-pollinated taxa with convergent anthers. It should be noted in this context that initially, i.e., immediately after the dehiscence of the pollen sacs, these windows are not yet obscured by the protruding stigmatic areas; the latter are still considerably smaller at this stage (flowers protandrous). It is, in our opinion, easy to imagine that a visiting insect can vibrate pollen out of the interior of the "anther-style and stigma complex."

Vibratile pollination may also work in taxa of subgenera *Phanerochiton* and *Dichroanthes*, both characterized by elongated stigmatic areas. It is possible that pollen is equally well "vibrated" out of the longitudinal slits between the anthers as out of the apical "windows" of the taxa of subgenus *Mitracme*.

As mentioned before, the floral features of *Acranthera* are diverse, and it is probable that the floral biology may be entirely different in some of the subgenera not investigated in detail.

Although characterized by anther-style and stigma complexes similar to those described above, the two "northwestern" subgenera, subgenus *Acranthera* (as "*Eu-Acranthera*" in Bremekamp, 1947; Sri Lanka and Indian Peninsula) and subgenus *Androtropis* (India (Assam) and peninsular Thailand), differ from other *Acranthera* taxa in some essential ways:
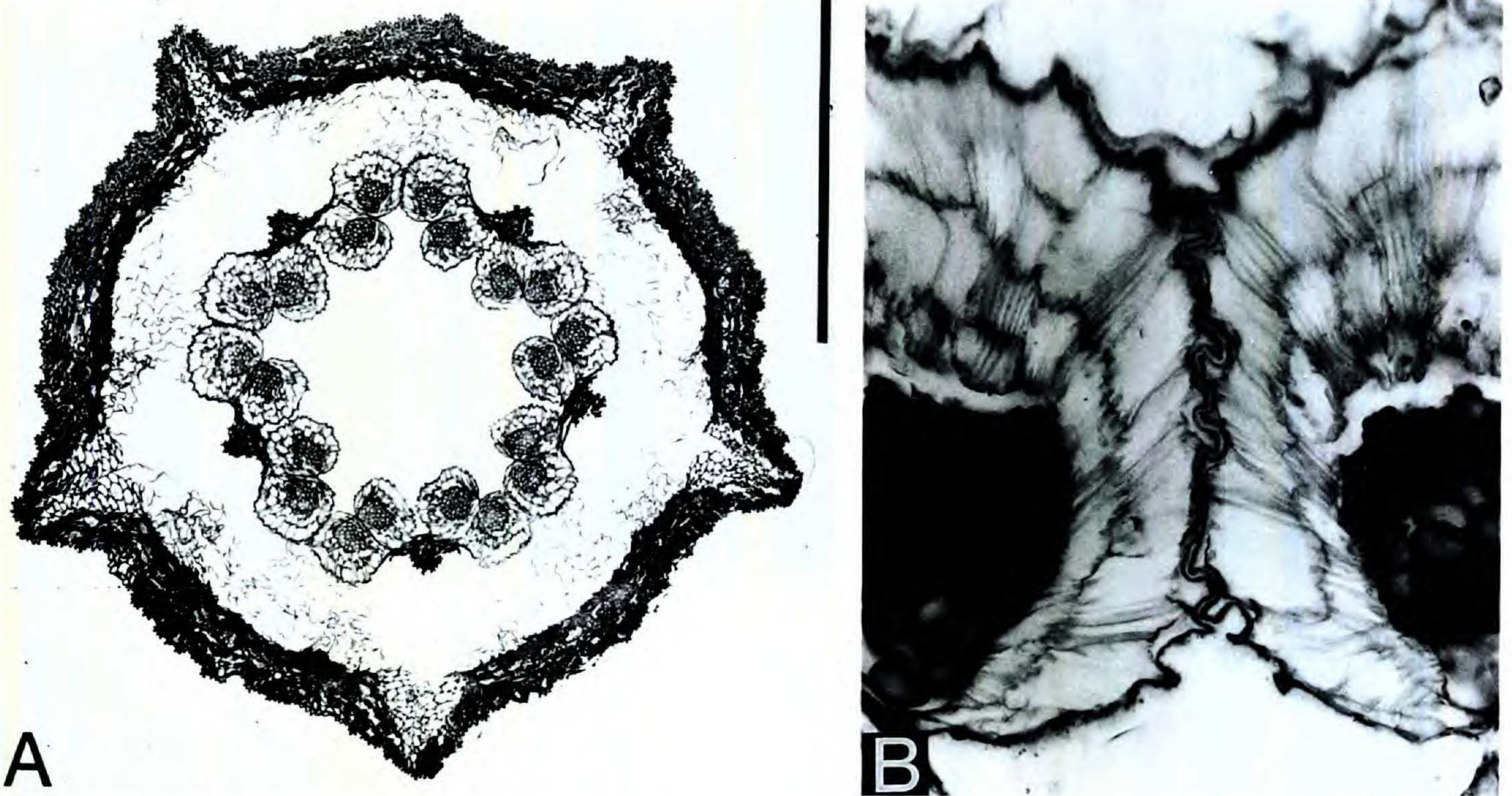
Figure 12. *Antherostele grandistipula.* —A. Cross section of corolla and anther tube.—B. Detail of A, showing agglutinate cuticles of pollen sacs from two neighboring anthers. Scale bar: 1 mm for A, 0.1 mm for B.

• All taxa of these two subgenera have a ringlike, nectar-producing disk, indicating nectar-collecting insects as pollinators. A disk, or nectar production for that matter, is absent in all other taxa of *Acranthera*; also cf. Bremekamp (1947).

• All representatives have flowers that are relatively much larger than in the other subgenera (corolla tubes ca. 20–40 mm long vs. hardly ever more than 10 mm), and the shape of the corolla tubes differs in being entirely infundibuliform or narrowly cylindrical below and infundibuliform above. Corolla colors—blue or violet in subgenus *Acranthera* and greenish to yellowish with pink spots inside in subgenus *Androtropis*—differ from those of the other subgenera.

• The filaments are very long (sometimes > 30 mm; several times longer than the anthers), whereas in other subgenera filament lengths rarely exceed 1 mm. In subgenus *Acranthera*, the filaments are free; in subgenus *Androtropis*, the lower half of the filaments is united into a filament tube which surrounds the style.

The two species Wong (1982) transferred from *Gardenia* to *Acranthera* belong to subgenus *Androtropis* (the distribution range of the subgenus is thus extended to include the Malay Peninsula). The illustration of one of them, *A. pulchella*, clearly shows the united filaments (Wong, 1982: fig. 3B), although this characteristic feature is not mentioned in the text.

Field observations of representatives of these taxa are needed.

ANTHEROSTELE (FIG. 12)

Bremekamp (1940) used the presence of an anther tube as one of the main characters separating *Antherostele* from *Urophyllum* and stressed that the former is the only genus in the *Urophyllum* alliance to show this phenomenon.

The genus has strictly cylindrical anther tubes. In contrast to, for example, *Argostemma* or *Steenisia* spp., apical connective appendages are not involved in the formation of the tubes. Minute apical connective appendages are present, but they—together with the very tips of the thecae—are curved outward. Except for the upper- and lowermost parts of the thecae, the anthers are united into a tube by agglutination of the cuticles of neighboring pollen sacs (Fig. 12).

This kind of anther tube formation is characteristic of the Asteraceae (Compositae; cf. Thiele, 1988). *Antherostele* appears to be the only rubiaceous genus in which the anther tube is formed this way.

From developmental studies of Asteraceae flowers it is known that the agglutination of the anthers occurs at a quite late stage of floral development (Thiele, 1988). This may also be the case in *Antherostele*, although developmental investigations could not be carried out (in available older bud
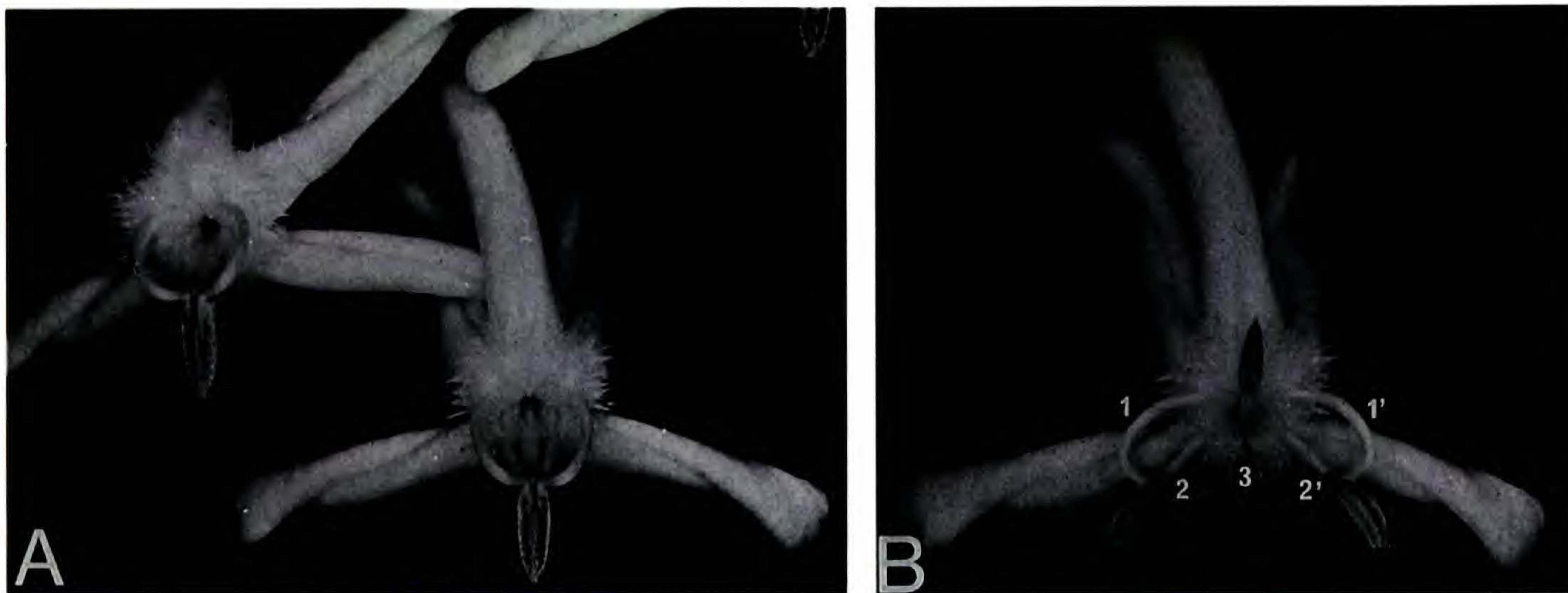
Figure 13. Flowers of *Posoqueria latifolia.* — A. Anthers still united. — B. Anthers separated: 2 pairs of lateral anthers (1 + 2; 1′ + 2′) and "lower" stamen (3) with its anther in an upper position (i.e., in front of the solitary, upper median corolla lobe; its filament blocks the entrance to the corolla tube).

stages, however, the anthers were already agglutinated).

The two species studied anatomically, *A. grandistipula* and *A. banahaensis*, show an identical situation. Judging from herbarium investigations, the two remaining species of the genus also do not differ in features of the anther tube (also cf. Bremekamp, 1940).

Comments on the pollination ecology.

Antherostele spp. are dioecious. The staminodes of female flowers, ca. half the size of the fertile stamens, are provided with completely sterile anthers which are also fused. In male flowers, the ovary is virtually absent, but a well-developed nonfunctional style is present. Whether the latter plays a secondary role in the pollination of *Antherostele* spp. is uncertain. It could serve as a receptacle for the pollen, which is released from the pollen sacs through inwardly directed longitudinal slits.

It is unlikely that in *Antherostele* the presence of an anther tube is linked with the occurrence of buzz pollination. Reasons for this tentative conclusion are:

● The genus's dioecy: Female flowers have relatively large, spreading stigma lobes that are exserted from the narrow corolla tubes and partially cover the corolla lobes. Because of the absence of the conspicuous stigma lobes, the appearance of the male flowers is different: the corolla lobes are fully exposed; the anther tubes are (except for the most apical parts) entirely included in the relatively long (normally > 10 mm) and narrow, cylindrical corolla tubes.
● The construction of the anther tubes themselves

(absence of apical constrictions; hardly like anther cones with apical, porelike openings).
● The presence of a ringlike nectar-producing disk on the roof of the inferior ovary.
● The dense indumentum on the inside of the corolla, obscuring the entrance to the interior of the tube.

Actual field observations are needed.

POSOQUERIA (FIGS. 13, 14)

In a widely overlooked article, Müller (1866) presented detailed information on the remarkable "catapult mechanism" of the flowers of *P. latifolia* (as "*Martha* (*Posoqueria?*) *fragrans*") by which pollen is hurled at the pollinator. Later, Hallé (1967) dealt with the "pollen projection" in this genus, unknowingly dealing with the same species as Müller (because he considered "*Posoqueria fragrans*" to be specifically distinct from *P. latifolia*), and with *P. longiflora.*

When a flower of *P. latifolia* opens, all anthers are united into an ellipsoid structure (Fig. 13A), which is in an oblique position in relation to the corolla tube (a reflection of the distinctly zygomorphic bud shape). Normally, the anther complex points downward (as shown by Müller, 1866: fig. 2 and in fig. 13A), but it was observed that even within a single plant, the united anthers may point upward as the entire flower is turned 180°. This needs to be mentioned, because all of Hallé's (1967: pl. 40, 2–6) drawings of *P. latifolia* show flowers in this more atypical inverted position, i.e., the slightly enlarged "upper" corolla lobe (cf. Fig. 13A; comparable to the "standard" of a leguminous flower) is directed downward.

The anther head is held a few millimeters away from the throat of the corolla by the unequal filaments and, notwithstanding its "down-" or "upward" position, it leaves free access to the entrance of the corolla tube. As a pollinator (hawkmoths according to Müller, 1886; subsequently confirmed by other authors, e.g., Bawa & Beach, 1983: table 1) approaches, it will try to insert its proboscis into the corolla tube. By doing so, it is likely to knock against the filaments 1 or 1′ (cf. Fig. 13). Touching the curved part of the filaments will trigger the mechanism and cause the anther head to separate explosively and to ejaculate the pollen. Müller, studying flowers at his home, noted that even flies are able to trigger off the catapult mechanism.

The anther complex breaks when triggered into three parts: two lateral pairs of anthers (Fig. 13B, 1 + 2, 1′ + 2′), which move outward due to an abrupt and rapid movement of the filaments, and the fifth anther (labeled "3" in Fig. 13B). As the anther complex separates, the filament of this anther curves up- or downward (depending on the orientation of a flower; cf. above) so that its broadened base "blocks" or obscures the entrance to the corolla tube (cf. Fig. 13B, filament "3"). After a while (8 to 12 hours according to Müller), this filament starts straightening. Presumably due to shrinkage processes the filament eventually recurves, thus again "unblocking" the entrance to the corolla tube. At this stage, the flower is functionally female (the two stigma lobes, hidden in the corolla tube, have unfolded and presumably are receptive). Hallé's (1967) statement (repeated by Robbrecht, 1988: 122) that the anthers are initially being held together only by an intermingling of their hairs is incorrect.

A combination of microtome and SEM investigations revealed the following situation for *P. latifolia*:

All anthers have both apical and basal appendages (Fig. 14A). The appendages are, in contrast to the other examples presented in this paper, not formed by the connectives but by sterile "extensions" of the thecae. Figure 14C, a section through the basal part of an anther complex, documents that 5 × 2 appendages are present, whereby two appendages each (extensions of the thecae of a single anther) are associated with a given filament. The contact area between appendages from *neighboring* anthers is easily recognized by the intensively (dark) stained epidermal layers. As shown in Figure 14D, the epidermis layers are only united by a fusion of their cuticles. Already Müller (1866) had indicated that, under the microscope, the anthers might be united by means of special "kitt."

In view of the later separation of the anther head into two anther pairs and a third anther, it should be noted that, at least in late bud stage, there appears to be no qualitative difference in the fusion of the five anthers. There is no obvious "stronger" fusion between the basal appendages of the stamens 1 + 1′ and 2 + 2′.

The pollen sacs dehisce longitudinally, and dehiscence takes place when the anther head is still intact (Fig. 14E). Consequently, an anther head in a stage as depicted in Figure 13A will contain a mass of pollen in its central hollow. The thecae of neighboring anthers themselves are not united (Fig. 14E). The close proximity of the pollen sac walls of adjacent anthers, nevertheless, effectively prevents pollen from reaching the exterior. The pollen grains, moreover, remain united in clumps (Fig. 14E; the presence of pollen kitt could, however, not be verified in the preserved material available). The later ejaculation of the pollen as "one spherical mass" (Hallé, 1967) is neither confirmed by personal observations by one of us (C. P.) nor by Müller (1866). The pollen is rather "shot" out of the exploding anther head in irregularly shaped clumps of different sizes (as is indicated by the section, Fig. 14E). Müller commented on the stickiness of these pollen clumps and noted they would even stick to smooth surfaces such as the blade of a knife.

Apically, each anther also has two sterile appendages, which are arranged on either side of the connective (Fig. 14F). Like the basal appendages, the apical ones, too, have an intensively stained epidermis, and in the most apical part the fused cuticles of neighboring epidermal layers ensure that the anthers are united (Fig. 14G). The area over which the apical appendages are united is very short. Less than half a millimeter from the very top, the appendages are already free. The section depicted in Figure 14F documents this. It is not at an entirely right angle to the anther head's long axis but slightly oblique. For this reason, only the apical appendages of the anther pair 1′ + 2′ are seen as being united.

The entire anther head, consequently, is a very "unstable" construction. Careful removal of the anther pair 1 + 2 from the anther complex depicted in Figure 14A, for example, showed that the apical appendage of anther 3 is hardly connected to the anther pair 1′ + 2′ (Fig. 14B; the arrows point to a distinct gap between the appendages). The fragile construction, however, is a prerequisite for the described catapult mechanism to work properly.

To date it is not known whether this unique pollen ejaculation mechanism occurs in all *Poso-*
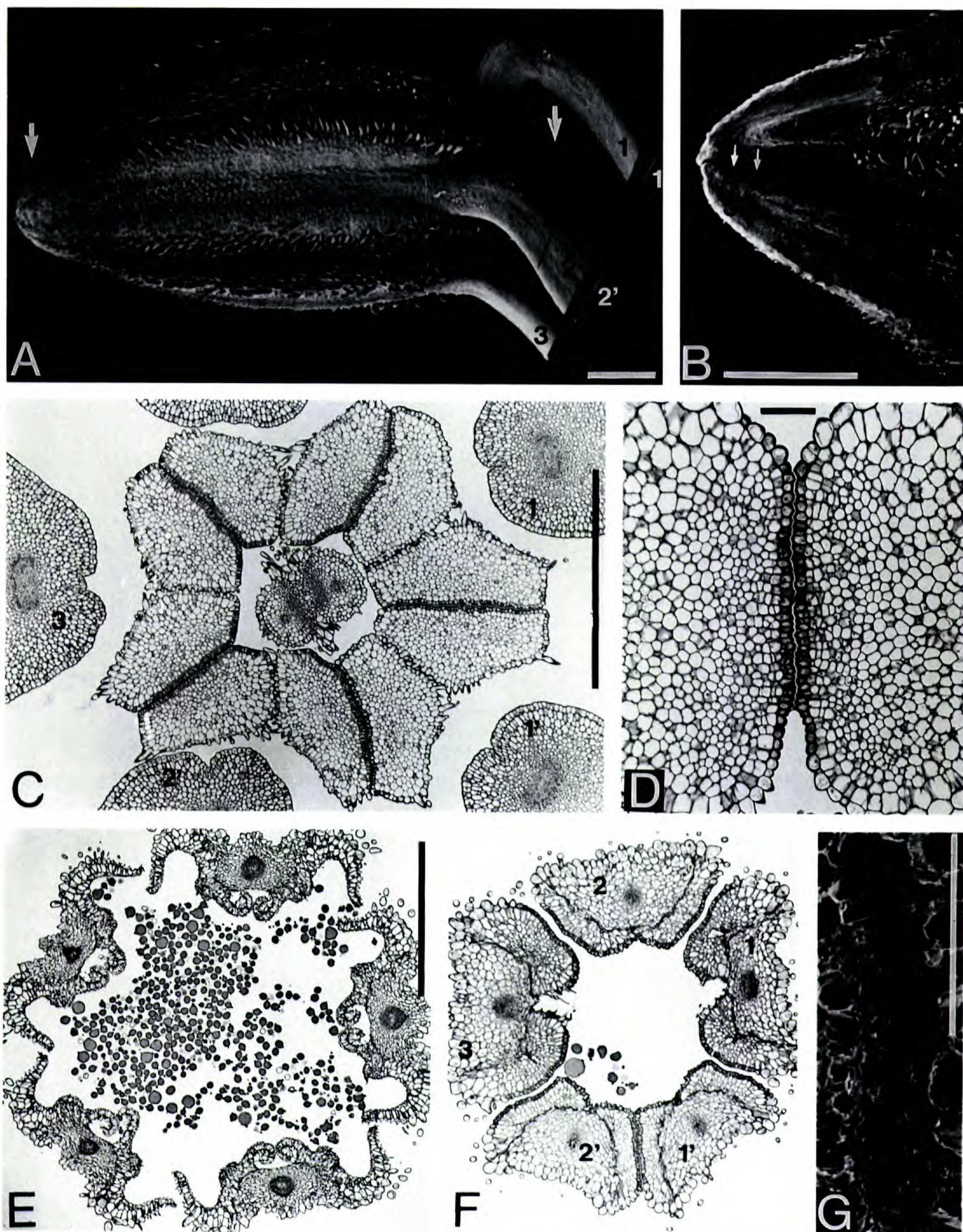
Figure 14.   Androecium of *Posoqueria latifolia* (stamens numbered as in Figure 13B).—A. United anthers, removed from bud shortly before anthesis; note apical and basal appendages (arrows).—B. Apical anther appendages, stamens 1 + 2 removed; note loose connection between anther 3 and anthers 1' + 2' (arrows).—C. Cross section of basal part of anthers; from outside in, filaments (numbered as in A), ring of basal anther appendages (10 paired sterile extensions = basal parts of the thecae), and two-lobed stigma (from bud; at later developmental stages, anthers and stigmas will be widely separated).—D. Detail of C; contact zone between sterile basal theca appendages of two neighboring stamens.—E. Cross section through the middle of an anther complex (from a flower in a stage as depicted in Fig. 13A, i.e., anthers still held together by apical and basal appendages), showing longitudinally dehisced pollen sacs and pollen mass in the central hollow.—F. Cross section of uppermost part of apical anther appendages; note that each connective has remnants of sterile thecae on either side; only appendages 1' + 2' are in direct contact,

queria species. Burck (1884) dealt with a "*Po-soqueria hirsuta*," which supposedly lacks this mechanism. The species is not included in Andersson's (1992) checklist of neotropical Rubiaceae, and Burck may have misidentified the plant investigated by him.

MISCELLANEOUS OTHER TAXA

Neurocalyx *Hook. (Argostemmateae)*.

The general floral morphology of this small genus (4 species; 3 in Sri Lanka, 1 in southern India) is similar to those *Argostemma* species with (sub)rotate corollas and anthers united into a tube. As in *Argostemma* spp., the androecium is comprised of introrsely dehiscent stamens with short, free filaments, and enlarged connectives are prominent on the outside of the anther tube; the pollen sacs open by means of longitudinal slits. Judging from Bremer's (1979) illustrations, agglutinated or fused apical connective appendages ensure the "stability" of the anther tube.

Corolla shape (starlike) and color (white; occasionally pink?) and yellow, exposed anther tube (cf. Bremer, 1984: fig. 1B, color illustration of *Neurocalyx gardneri*) suggest that *Neurocalyx*, from the context of pollination biology, is inseparable from numerous *Argostemma* species. The genus can safely be added to the group of buzz-pollinated Rubiaceae.

Siphonandrium *K. Schum. (incertae sedis)*.

This is a poorly known, dioecious New Guinean monotypic genus (*S. intricatum* K. Schum.) of uncertain taxonomic position (possibly belonging to the Morindeae s. str.; cf. Igersheim & Robbrecht, 1993). It appears to be unique in the Rubiaceae in that not only an anther but also a filament tube is present, i.e., a staminal tube. Quoting Schumann (1905), "The filaments are tightly fused and the anthers are glued together . . . so that one has the impression of only one anther being present." According to Schumann, the staminal tube (> 4 mm long) is exserted from the hypocrateriform corolla; the latter is characterized by a short tube (3 mm long) and relatively large, spreading lobes (5 mm long); no information on anther dehiscence is included. Based on these data, the possibility of buzz pollination cannot be excluded altogether.

No material was seen; verification is badly needed.

Molopanthera *Turcz. (Cinchoneae; provisionally transferred to Condamineeae by Andersson & Persson, 1991)*.

According to Schumann (1891: 10, 54), the androecium of this monotypic eastern Brazilian genus resembles that of *Posoqueria* "in all details." Like *Posoqueria*, it has zygomorphic flowers, and Schumann suggested that it "most probably" exhibits the same mechanism of explosive pollen release.

No material was seen; verification is needed.

Two more genera of the Condamineeae deserve mentioning: Tresanthera *Karst. and* Rustia *Klotzsch.*

In conjunction with buzz pollination, both genera are included in Buchmann's (1978) table 5. *Tresanthera* flowers have anthers in a tube- or conelike arrangement (whether actual agglutinations or fusions are present remains unknown). The apically inwardly curved anthers open by means of a subapical valvelike structure. Anther morphology seems to be comparable to that of *Argostemma neurocalyx* (cf. Fig. 6C) except that the opening mechanism of the pollen sacs is apparently more specialized. Floral structure definitely suggests the occurrence of buzz pollination.

In contrast, the anthers of *Rustia*, dehiscing with two apical pores, do not form an anther cone. The genus's flowers do not fall into the "*Solanum*-type" of pollen flowers category, although the occurrence of buzz pollination cannot be excluded altogether.

Actual field observations on taxa of both genera are encouraged.

DISCUSSION

The investigations presented here reveal the diversity by which "synandry" (in a very wide sense) is achieved in genera and species of the Rubiaceae. It is remarkable that even within genera (*Argostemma*) there are considerable differences in detail (Table 1).

The occurrence of this phenomenon is scattered

←

as the section is slightly oblique rather than strictly perpendicular to the anther complex's long axis.—G. Contact zone between apical sterile theca appendages of two neighboring stamens from near the very apex of the anther complex. A, B, G: SEM-graphs; all others: LM-graphs. Scale bars: 1 mm (A; B; C = F; E); 0.1 mm (D; G).

Table 1. Summary of the androecium features, the type of anther dehiscence, and the presumed pollination mode of the investigated Rubiaceae.

Androecium	Anther dehiscence	Pollination	Taxa	Position
Anther tube present				
(1) Tube apically constricted, with porelike opening				
(1a) Apical connective appendages interlocked and agglutinated	longitudinal slits	buzz pollination	*Steenisia*	Rondeletieae
(1b) Apical connective appendages united, in part also pollen sac walls agglutinated	longitudinal slits	buzz pollination	*Argostemma* p.p.; *Neurocalyx* (?)	Argostemmateae
(2) Tube not apically constricted, strictly cylindrical; pollen sac walls agglutinated; connective appendages small, not united	longitudinal slits	?	*Antherostele*	Urophylleae
(3) Tube with partially united, crownlike apical connective appendages; anthers united by means of special cell layer	poricidal	buzz pollination	*Strumpfia*	incertae sedis
Anthers not united				
(1) Anthers long and narrow, connivent	poricidal	buzz pollination	*Argostemma* p.p. (e.g., *A. diversifolium*)	Argostemmateae
(2) Anthers flattened, in ± zygomorphic arrangement	poricidal	buzz pollination (?)	*Argostemma* p.p. (*A. neurocalyx*)	Argostemmateae
"Anther-style and stigma complex"				
Apical connective appendages and stigma apex united; in part formation of apical "windows" (= gaps between connective appendages)	longitudinal slits	buzz pollination (?)	*Acranthera* (p.p.)	Isertieae ?; incertae sedis
Anther head				
Basal and apical theca appendages united	longitudinal slits	"catapult mechanism"; hawkmoth	*Posoqueria* (also *Molopanthera* ?)	Gardenieae subtribe Gardeniinae (Condamineae)

and occasional in the family. With the exception of the undoubtedly closely allied species pair *Argostemma* and *Neurocalyx*, it does not seem to reflect relationships. Its presence must rather be seen as a pollination biological adaptation or specialization that has arisen repeatedly in the family.

Among the Rubiaceae with united anthers, the group of buzz-pollinated "*Solanum*-type" of pollen flowers is by far the most prominent.

In the Rubiaceae, a family in which the "basic" type of pollen sac dehiscence is by means of longitudinal slits (cf. Robbrecht, 1988: 86), the poricidally dehiscent anthers of buzz-pollination systems are mostly "replaced" by an alternative construction, i.e., solid anther tubes (with longitudinally dehiscent pollen sacs); see under *Argostemma* for details. Yet, some buzz-pollinated Rubiaceae have also evolved poricidally dehiscent buzz anthers (whereby this is not necessarily linked with the union of anthers into solid tubelike structures).

Argostemma is the only Rubiaceae genus known to date in which both anther tubes with longitudinally dehiscent pollen sacs and free anthers with apical pores are found. In addition to this "switch" to poricidal dehiscence, possibly indicating a more "advanced" state, *Argostemma* is also remarkable because of the repeated occurrence of zygomorphy both in androecium and corolla.

Apart from the very specialized, monotypic *Strumpfia* (cf. Igersheim, 1993a), other genera with anthers with apical pores are *Rustia*, *Tresanthera*, and *Stomandra*, all belonging to tribe Condamineeae. Detailed morphological and anatomical information on their stamens is, however, still lacking. At present it is not known whether the "apical pores" of their anthers are in fact homologous to "normal" apical pores as in, for example, *Argostemma diversifolium*, or are very specialized modifications.

The unique anther-style and stigma complexes of *Acranthera* spp. with their peculiar "windows" (gaps between the elongated apical connective appendages) may represent a hitherto totally unknown "alternative" construction of a buzz-pollinated blossom. Actual proof from field observations is lacking, however. In this context it must be stressed again that the amount of in-depth field studies on taxa of the Rubiaceae, in general, is dismally small. More emphasis on this kind of work is desperately needed.

Another example of the lack of field data and pollination ecological information is *Antherostele*. The morphological and anatomical evidence presented here, together with the investigation of herbarium material, does not provide adequate clues to the possible pollination mechanism. We are inclined to believe that it might not be buzz pollinated.

From the viewpoint of pollination ecology, *Posoqueria*—with its unique explosive pollen release mechanism—stands apart from the remainder of the investigated genera and species; its pollination by hawkmoths has been confirmed independently by several authors. The presented data, nevertheless, provide the anatomical explanation for the temporary union of their anthers and correct previously published information.

The pollination mechanism of the investigated *Posoqueria* spp. may or may not cast some doubt on the taxonomic position of the genus in the Gardenieae–Gardeniinae. The tribe is characterized by the presence of an "ixoroid mechanism," i.e., secondary pollen presentation by the style and/or outside of the stigma lobes (Robbrecht & Puff, 1986), but in the investigated *Posoqueria* spp. this feature is absent and, in fact, must be absent if the mechanism of pollen release is to function properly. The pollination biological specialization could be interpreted as being an aberrant feature, and the genus left in the Gardenieae. On the other hand, pollination mode, or the characteristics that go with it (e.g., zygomorphy of the corolla), could be taken as strong indications for a position outside the Gardenieae. Detailed investigations of additional character states should help in clarifying its position.

Literature Cited

Andersson, L. 1992. A provisional checklist of neotropical Rubiaceae. Scripta Bot. Belg. 1: 1–199.
———— & C. Persson. 1991. Circumscription of the tribe Cinchoneae (Rubiaceae)—A cladistic approach. Pl. Syst. Evol. 178: 65–94.
Bawa, K. S. & J. H. Beach. 1983. Self-incompatibility systems in the Rubiaceae of a tropical lowland wet forest. Amer. J. Bot. 70: 1281–1288.
Bremekamp, C. E. B. 1940. *Antherostele*, genus novum Rubiacearum *Urophyllo* affine. J. Arnold Arbor. 21: 25–31.
————. 1947. A monograph of the genus *Acranthera* Arn. ex Meisn. J. Arnold Arbor. 28: 261–308.
Bremer, B. 1979. The genus *Neurocalyx* (Rubiaceae-Argostemmateae) in Ceylon. Bot. Not. 132: 399–407.
————. 1984. The genus *Steenisia* (Rubiaceae) and its taxonomic position. Nordic J. Bot. 4: 333–345.
————. 1989. The genus *Argostemma* (Rubiaceae-Argostemmateae) in Borneo. Ann. Missouri Bot. Gard. 76: 7–49.
Buchmann, S. L. 1978. Vibratile ("buzz") pollination in angiosperms with poricidally dehiscent anthers. PhD. Thesis, University of California, Davis.
————. 1983. Buzz pollination in angiosperms. Pp. 73–113 *in* C. E. Jones & R. J. Little (editors), Handbook of Experimental Pollination Biology. Van Nostrand Reinhold, New York.

Buchner, R. & C. Puff. 1993. The genus complex *Danais–Schismatoclada–Payera* (Rubiaceae). Character states, generic delimitation and taxonomic position. Bull. Mus. Natl. Hist. Nat., Sér. 4, B, Adansonia, 15(1–4): 23–74.

Burck, M. W. 1884. Sur l'organisation florale chez quelques Rubiacées. Ann. Jard. Bot. Buitenzorg 4: 12–87.

Goebel, K. 1923. Organographie der Pflanzen, insbesondere der Archegoniaten und Samenpflanzen. Zweite, überarbeitete Auflage. Dritter Teil: Spezielle Organographie der Samenpflanzen. Fischer, Jena.

Hallé, F. 1967. Étude biologique et morphologique de la tribu des Gardeniées (Rubiacées). Mém. O. R. S. T. O. M. 22: 1–146.

Igersheim, A. 1993a. The character states of the Caribbean monotypic endemic *Strumpfia* (Rubiaceae). Nordic J. Bot. 13: 545–559.

——. 1993b. Gynoecium development in Rubiaceae–Vanguerieae, with particular reference to the "stylar head"-complex and secondary pollen presentation. Pl. Syst. Evol. 187: 175–190.

—— & E. Robbrecht. 1993. The character states and relationships of the Prismatomerideae (Rubiaceae–Rubioideae). Comparisons with *Morinda* and comments on the circumscription of the Morindeae s.str. *In:* E. Robbrecht, Advances in Rubiaceae Macrosystematics. Opera Bot. Belg. 6: 61–79.

Müller, F. 1866. Über die Befruchtung der *Martha (Posoqueria?) fragrans*. Bot. Zeitung (Berlin) 23: 129–133.

Puff, C. & D. E. Mantell. 1982. The tribal position and affinities of *Crocyllis* (Rubiaceae). Bot. Jahrb. Syst. 103: 89–106.

——, L. Andersson, U. Rohrhofer & A. Igersheim. 1993. The tribe Schradereae (Rubiaceae) reexamined. Bot. Jahrb. Syst. 114: 449–479.

Robbrecht, E. 1988. Tropical woody Rubiaceae. Opera Bot. Belg. 1: 1–271.

—— & C. Puff. 1986. A survey of the Gardenieae and related tribes (Rubiaceae). Bot. Jahrb. Syst. 108: 63–137.

Sazima, M., S. Vogel, A. Cocucci & G. Hausner. 1993. The perfume flowers of *Cyphomandra* (Solanaceae): Pollination by euglossine bees, bellows mechanism, osmophores, and volatiles. Pl. Syst. Evol. 187: 51–88.

Schumann, K. 1891. Rubiaceae. Pp. 1–156 in A. Engler & K. Prantl (editors), Die natürlichen Pflanzenfamilien 4(4). Engelmann, Leipzig.

——. 1905. *Siphonandrium*. Pp. 394–395 in K. Schumann & K. Lauterbach, Nachträge zu Flora der deutschen Schutzgebiete in der Südsee (mit Anschluß Samoa's und der Karolinen). Gebrüder Borntraeger, Leipzig.

Teppner, H. 1993. Die Tomate—Verwandtschaft, Geschichte, Blütenökologie. Pp. 189–211 in G. Aubrecht, M. Brands, F. Gusenleitner, F. Speta & S. Weigl (editors), Amerika—Zur Entdeckung—Kulturpflanzen—Lebensraum Regenwald. Kataloge des OÖ. Landesmuseums, Neue Folge 61.

Thiele, E.-M. 1988. Bau und Funktion des Antheren-Griffel-Komplexes der Compositen. Diss. Bot. 117: 1–171.

Vogel, S. 1978. Evolutionary shifts from reward to deception in pollen flowers. Pp. 89–104 in A. J. Richards (editor), The Pollination of Flowers by Insects [Linnean Society Symposium Series 6]. Academic Press, London.

Wong, K. M. 1982. Notes on *Gardenia* and *Acranthera* (Rubiaceae) from Peninsular Malaysia. Gard. Bull. (Singapore) 35: 21–32.